USE OF FILTRATE FROM CROP RESIDUE ASH FOR COOKING IN RURAL HOUSEHOLDS IN NORTHERN UGANDA

by

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Abstract

Filtrate made from crop residue ash is used as a traditional cooking additive in Northern Uganda to decrease cooking time of legumes and impart a culturally preferred taste. The environmental and health implications of this practice are poorly understood. Research conducted in Northern Uganda and Canada included comparison of cooking times for black beans (*Phaseolus vulgaris* L.) among four cooking water treatments, yielding an 18% time savings with ash filtrate over the control. Palatability evaluation revealed beans cooked with ash filtrate were not preferred over other treatments. Study site and treatment significantly impacted overall palatability scores. Inductively coupled plasma - mass spectrometry analysis showed high mineral content for crop ash but low values for crop ash filtrate. Daily consumption amounts of beans cooked with ash filtrate contributes elemental levels (e.g., calcium and iron) within nutritional standards. However, the highly alkaline nature of ash filtrate may have anti-nutritional effects on diet.

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Acknowledgements

This thesis would not have been possible without the women and men I had the great fortune to learn from in Northern Uganda. I was graciously accepted into homes and communities to study a practice integral to their culture, and for this experience I am so grateful. The strength and work ethic of the Ugandan women I had the privilege to meet will stay with me forever. Special thanks to Geoffrey Odongo and Anna Akello for facilitating my research while in Uganda.

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Chapter 1. General introduction

In rural Northern Uganda, there is a prevalent indigenous practice of leaching water through ash material generated from crop plant residue to obtain a concentrated ash filtrate. This ash filtrate is added to cooking water of specific foods, such as hard-to-cook dried legumes. Local women in the region have expressed two main anecdotal reasons for this practice. First, it is believed that plant ash filtrate shortens the cooking time of several staple foods, and second, it is thought that use of the filtrate improves the palatability of certain foods. Beans and other legumes are staple foods in developing countries, including Uganda (Uzogara, Morton, & Daniel, 1990), but are notoriously difficult to cook, especially after undergoing a 'hardening' stage, a physiological consequence of poor storage conditions (de León, Elias, & Bressani, 1992; Reyes-Moreno & Paredes-Lopez 1993).

Various plant ash samples contain elevated levels of sodium, potassium and other elements, and are highly alkaline in nature (Calloway, Giauque, & Costa, 1974; Kuhnlein 1980; Ohtsuka, Suzuki, & Morita, 1987; Kaputo 1996; Wanjekeche, Wakasa, & Mureithi, 2003; Mamiro *et al.* 2011). Cooking time of dried beans is reduced by addition of sodium to cooking water (Varriano-Marston & de Omana 1979; Van Buren 1986), as well as by increased alkalinity (Ankrah & Dovlo 1978; Uzogara *et al.* 1990; Onwuka & Okala 2003; Wanjekeche *et al.* 2003). Anecdotal beliefs support these findings, as plant ash filtrate is regularly added to cooking water to decrease cooking time. As most cooking in rural areas of East Africa still takes place over a wood fire (Rubaihayo 1995; Tabuti, Dhillion, & Lye, 2003), the length of cooking time has a direct impact on both the local environment (e.g., fuel wood use and supply) and associated daily activities (e.g., time spent collecting fuel wood and time spent preparing meals). In addition to reduced cooking time, ash filtrate is thought to give foods a distinct flavour and smell which has become culturally important over time.

Safe water and food sources are the most important influences on an individual's opportunity for a productive and successful existence (Rosegrant & Cline 2003). Unfortunately, due to a variety of political and economic reasons these are often compromised in developing areas and/or countries. The Food and Agriculture Organization (FAO) of the United Nations defines food security as 'a situation that exists when all people, at all times, have physical, social and economic access to sufficient, safe and nutritious food that meets their dietary needs and food preferences for an active and healthy life' (FAO 2008). From this description, four key dimensions are identified: physical availability, economic and physical access, use and stability of each of the previous dimensions over time (FAO 2008). The World Health Organization is closely aligned with the FAO as it identifies food availability, food access and food use as necessary for a healthy existence (WHO 2012).

This study focuses on the food utilization aspect of food security, inclusive of food practices, preparation, diversity and distribution within the household (FAO 2008). More specifically, this study examines whether a traditional practice of food preparation contributes to human health problems. In rural areas, especially those in developing countries, there is often a delay in technological and/or infrastructural advancement. This is due to geographical factors (e.g., dispersed population base), economic factors (e.g., lack of funding), and political factors (e.g., past conflict in Northern Uganda) which have hindered program establishment. This lack of outside information and/or opportunities for formal education makes the oral practice the primary process of knowledge transfer within communities. For these reasons, residents in remote areas rely heavily on traditional practices in their daily activities, such as the methods used in food preparation.

Other authors have examined the use of this practice in Africa (Huntingford 1955; Whitby 1972; Onwuka and Okala 2003), studied the effect of ash or ash filtrate use on cooking time (Wanjekeche *et al.* 2003; Mamiro *et al.* 2011) and sensory properties (Wanjekeche *et al.* 2003), and analyzed the elemental composition of plant ash (Kuhnlein 1980; Kaputo 1996; Mamiro *et al.* 2011). However, there are no studies documenting the production of plant ash filtrate, or examining the practice in its entirety. A distinct gap in knowledge lies in examining the potential health effects of using this filtrate for cooking and therefore, in consumption. This study draws an interdisciplinary link between the perceived benefits of using plant ash filtrate for cooking and the potential health and environmental implications. The outcome of this research will be used to inform policy recommendations and develop educational opportunities around the use of ash filtrate as a cooking additive, with the intent of improving lives in rural Northern Uganda and other places where this practice is used.

1.1. Organization of thesis

This thesis is structured in a manuscript form with separate chapters prepared for submission for publication in a peer-reviewed scientific journal. Chapter one provides an overall introduction to the study topic. Chapter two gives contextual background information on Northern Uganda and the study site communities, as well as a brief account of the use of traditional salts used in rural Uganda. The second chapter also provides a description of research materials collected from the communities, with a full description of the generation of ash filtrate. Chapter three gives an overview of previous research done on relevant cooking additives. Chapter four pertains to the first objective of this research, which is the comparison of cooking times among study treatments. Chapter five focuses on the second and third objectives which include several analyses of palatability (sensory) preferences, with respect to the treatments. Chapter six pertains to the last objective and details the composition of the plant ash and filtrate, with specific attention given to identifying any injurious properties or outcomes resulting from its use. Chapter seven provides a summary of findings, identifies policy and educational suggestions, and provides recommendations for future research opportunities.

Chapter 2. Background information and general research premises

2.1 Northern Uganda

2.1.1 History

Uganda has an interesting and turbulent history. Until 1893 Uganda had been ruled by several kingdoms. In 1894, the British army colonized by force the area now known as Northern Uganda, and Uganda became a protectorate of the British Empire. During this time, it was famously coined the 'Pearl of Africa' by Sir Winston Churchill for its extraordinary environmental and cultural diversity and beauty. In 1962, Uganda gained independence from Britain and has since witnessed a tragic history of violence and unrest, most notably under the rule of Idi Amin Dada. Recent conflicts in the northern part of the country between government forces and Joseph Konys' Lord's Resistance Army (LRA) ended in 2006. Uganda is making great progress in returning to a stable and progressive country.

2.1.2. Demographics and socio-economic factors

There are four administrative units in Uganda: Northern, Western, Eastern, and Central, each of which are broken down further into districts, counties, sub-counties, parishes, and villages (UBOS 2012). District boundaries have rapidly changed in the past few years for a present total of 111 districts plus Kampala, the capital city (Figure 1). The current population estimate for Uganda is 34.1 million, and 7 million for all of Northern Uganda (UBOS 2012). The population is rapidly growing; Uganda has the third highest birth rate and fifth highest growth rate in the world at 3.32%, with an average of over 6 children per woman (CIA 2012). At the same time, Uganda has a very low life expectancy of only 52.65 years for males and 55.35 years for females due to a number of health and lifestyle issues. Prevalent health problems in the region include malaria, tuberculosis, HIV/AIDS, diabetes, and cardiovascular disease (UBOS 2012).

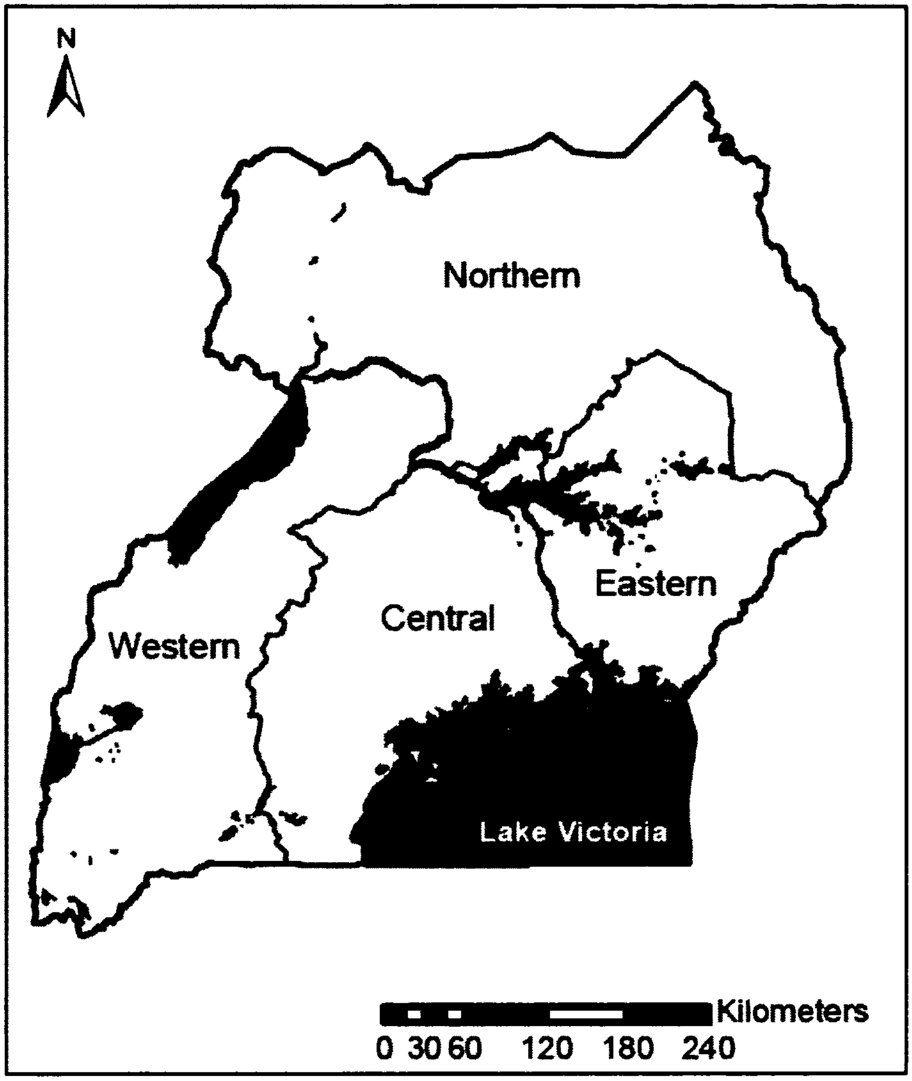

Figure 1. Country of Uganda, divided by administrative units.

Uganda is predominantly rural, with approximately 13% of citizens residing in urban centers. The majority of the urban population resides in Kampala and surrounding area (CIA 2012). Northern Uganda consists of mainly peasant farmers who rely on agriculture for their livelihoods. Average wages for general or agricultural labor are somewhere between 70,000 and 90,000 Ugandan Shillings (USh) or approximately $35.00 Canadian dollars (Cdn) per month (UBOS 2009). This estimate is much higher than the wages earned by many peasant farmers, based on socio-economic information collected in this study. Many families subsist solely on their own produce from harvest or goods from their farm animals. Depending on the crop yield, farmers can use opportunistic sale of produce at market for clothing, children's education, and other necessities. Additional household income through sale of produce and other agricultural goods is largely dictated by the time of season as dry periods can be severely limiting in most areas. Irrigation is scarce and a large proportion of farming is still done by hand (hoe). More affluent farmers use oxen or other animals.

Official languages of Uganda include English (due to British colonization until 1962) and Swahili (a regionally important business language). However, more than 50 languages are currently spoken throughout the country (UBOS 2012).

2.1.3. Climate and geography

Uganda is a land-locked equatorial country, sharing a border with five other East African countries: the Democratic Republic of the Congo, Sudan, Kenya, Tanzania and Rwanda. Average daily temperatures in Northern Uganda range from 20°C - 30°C year round and two rainy seasons occur from March-May and September-November. On average there is 1440mm of precipitation per year around the Lira area (UBOS 2012). The main sources of water include an arm of the nearby River Nile and Chobe River for agricultural purposes, while natural springs and boreholes provide water for household use (UBOS 2012). The predominant soil type in the region is a ferrisolic sandy clay (Ollier 1959), now referred to as an oxisol or ferralsol (Eswaran, Almaraz, van den Berg, & Reich, 1997; ISRIC 2013). These red soils are characterized by high iron and aluminum sesquioxides with subsequent high phosphorus retention but low available water holding capacity, and poor overall productivity. They are highly weathered and acidic soils supporting savannah and woodland type vegetation (UBOS 2012). Common crops in this region include bananas, legumes, sesame (*sim-sim*), maize, and sweet potatoes (Rubaihayo 1995).

2.2 Study site communities and qualitative methodology

The field-based portion of this research took place at four study sites in Northern Uganda; three in Oyam District, and one in Lira District (Table 1). Study sites were located as follows: Site 1 in Dog Abam Village (36N 426804m E, 246337m N), Site 2 in Telela Village (36N 492122m E, 243647m N), Site 3 in Arok Village (36N 426493m E, 245231m N), and Site 4 in Tit Village (36N 420040m E, 252369m N) (Figure 2). The choice of study sites was based on previous interaction between a non-governmental organization, the Northern Uganda Development Foundation (NUDF), and local community members. NUDF is a Canadian-based organization with field operations in Northern Uganda. Given the cultural and linguistic challenges, utilizing study sites with established relations to NUDF provided a base from which to begin to ensure a timely method of study.

Table 1. Study area classification and population figures collected from the Local Chairperson in each Parish.

Study Site	Village	Parish	Sub-County	County	District	Parish Population
1	Dog Abam	Kamdini	Kamdini	Oyam South	Oyam	6412
2	Telela	Boroboro	Adekokwok	Erute South	Lira	32,200
3	Arok	Akaka	Aber	Oyam South	Oyam	9960
4	Tit	Juma	Kamdini	Oyam South	Oyam	7999

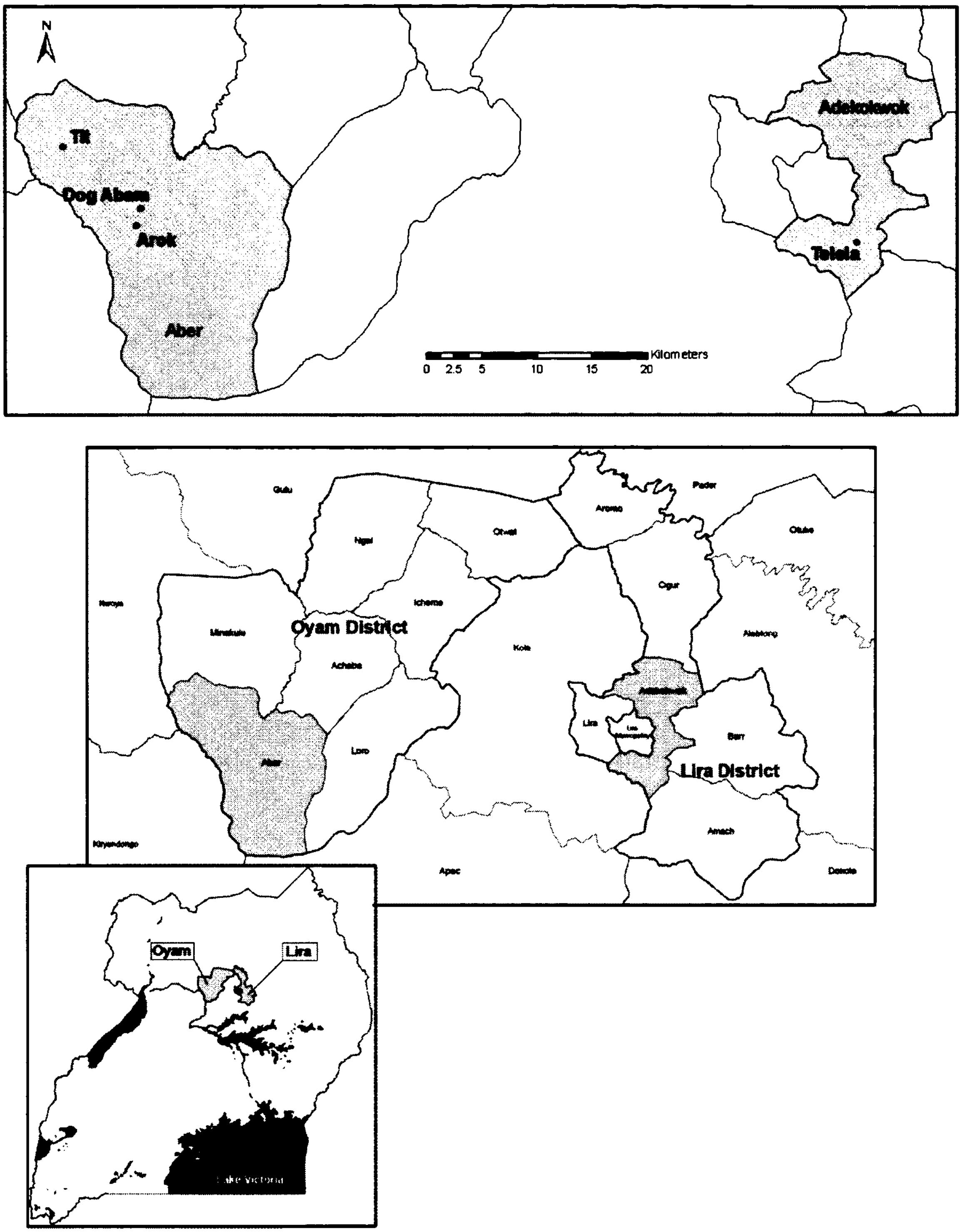

Figure 2. Relative location of study sites in Oyam and Lira Districts, Northern Uganda.

Oyam and Lira Districts are home to the Lango and Acholi; settlers originating from the area that is now South Sudan (UBOS 2012). The dialect spoken in the study area is Luo, of the Nilotic languages. General population information was obtained from the Local Chairperson (LC) (a local government representative), for each parish (Table 1). The 2012 projected population for Oyam District was around 350,000, and for Lira District was just over 650,000 (UBOS 2012). Oyam District has only one hospital and 15 health centres, serving a population of 350,000 people, while Lira District has two hospitals and 37 health centres which serve a population of 650,000 people (UBOS 2012). As such, access to health care can be difficult, evidenced by a statistical average of only 0.5 visits per person per year (or one visit every two years by a single person) to any formal health facility in Oyam District (UBOS 2012). This represents one of the lowest per capita rates of health care visits in Uganda. Poverty, inefficiencies in service, poor access, and cultural compatibility issues continue to place primary health care responsibilities on traditional healers instead of trained physicians (Kamatenesi, Acipa, & Oryem-Origa, 2011).

The lack of stable infrastructure, including health facilities, schools, and home settlements is largely due to the destructive LRA conflict in this area which took place from the mid-1980s until 2006. Nearly two million people were forced to flee from this region to internally displaced person (IDP) camps and remained there for a decade or more (UNHCR 2007). Upon returning they discovered homes, land, and communities which were no longer functional.

Rural areas, especially those in developing countries, are often the last to receive technological and/or infrastructural advancements. The difficulty in program establishment, lack of funding and a dispersed population base are some of the factors which impede progress. It is in these remote areas that traditional knowledge is highly relied upon. A lack of outside information, and/or formal education, makes the passing on of traditional knowledge a very important process within these communities. Domestic knowledge, including cooking practices, is usually disseminated by women to daughters or other female relatives (Madge 1994). This was confirmed to be the case for the use of ash filtrate as a cooking additive in Northern Uganda. All interview respondents identified either their mother or grandmother as the person who taught them about this practice. All respondents also identified this practice as a very important part of their life and culture, and many of these women said it was necessary for cooking properly. There can be resentment felt by older women when the next generation is not interested in learning these practices, as it is believed that traditional foods and cooking methods are healthy and will provide what is needed to survive (Kuhnlein, Calloway, & Harland, 1979).

2.2.1. Observation period and interviews

Through collaboration with NUDF and local women, a combination of qualitative and quantitative methods was used to gain a comprehensive understanding of this practice. In Northern Uganda methods used to collect data included semi-structured interviews, video and photo documentation of all ash and ash filtrate procurement procedures (from crop harvest through use in cooking), palatability tests, sample collection, and preliminary cooking time trials. Approval for all activities involving study participants was obtained from the Research Ethics Board at the University of Northern British Columbia (UNBC) prior to initiation of study. The interviews included both qualitative and quantitative questions covering aspects of the history, methods and use, reasoning, and perceptions about benefits and health effects of using the ash filtrate, as well as basic socio-economic information (Appendix A). Semi-structured interviews were chosen because of the adaptability during implementation (Mikkelsen 1995), allowing for the best information to be obtained about this unfamiliar practice. While the initial intention was to interview randomly selected women from within each district, it became apparent that lack of census data on citizens would prevent any type of random choice of informants. Instead my local research assistant/translator (Ugandan female, aged 40), who was hired to assist with field work, acted as a key informant, providing introductions to women representing a variety of demographics. Prior to data collection, I met with my local research assistant/translator to explain the process and goals of this study, answer any questions, and ensure privacy for all participants through a signed confidentiality agreement (Appendix B). I conducted two trial interviews to clarify and assess appropriateness of questions, determine length of interviews and gain familiarity in working with my research assistant. All interviews were conducted via translation at the homes of each respondent and were tape recorded if the environment allowed. In most cases it was decided that the background noise did not facilitate voice recording and answers were transcribed on paper. Incorporating qualitative questions allowed for a broader range of information to be gained about this relatively unknown practice. Understanding what this traditional practice means to the women of Northern Uganda will help inform any future policy and educational decisions. The quantitative questions added to a general understanding of this practice, as well as provided descriptive statistical support to each study objective. Information gathered through observation and interviews was applied to each objective of this study. The other methods of data collection conducted in the field and in Canada are fully explained in Chapters 4-6.

2.3. History and importance of indigenous salt use in rural Uganda

Salt is one of the oldest commodities known to human civilization, ubiquitous and used for both seasoning and preservation of food. Composed primarily of sodium chloride ($NaCl$), a certain amount of salt in our diet is required and there are several types available for human use. Commercial (table) salt is a refined and often iodized salt, where most of the impurities have been removed. Indigenous (ground) salt refers to a traditional type salt that contains both $NaCl$, as well as a mixture of additional minerals and other impurities (Townsend, Liao, & Konlande, 1973; Makanjuola & Beetlestone 1975; Kuhnlein 1980). Until relatively recent advances in refinement and transportation, this was the type of salt most relied on across the world. Ground salt is obtained most often as a precipitate from saline water sources, in a granular or rock form. There are several sources proximate to Northern Uganda capable of producing ground salt; Lake Magadi, Kenya; Lake Katwe, Uganda; Lake Natron, Tanzania; and El-Atrun, Sudan (Nielsen 1999; Nielsen & Dahi 2002). Collectively, this ground salt is referred to as '*magadi*' in eastern Africa, a salt which is usually scooped from the topmost layer of earth and which can also contain a mixture of other elements and soil (Nielsen 1999). Aside from personal use, ground salt is also transported to various town markets for sale by volume.

In communities that were located too far from ground salt sources and/or did not trade for salt, a lesser known traditional practice was used to obtain an indigenous 'salt' cooking additive from plants or plant parts. Plants or individual plant parts were burned for their ash, which was then added to food directly as ash, or used as a filtrate produced from the ash. This practice is still occurring in Uganda, and purportedly in several other African countries, Oceania, and South America (Ohtsuka *et al.* 1987; Kaputo 1996; Sharland 1997; Onwuka & Okala 2003; Wanjekeche *et al.* 2003; Mamiro *et al.* 2011; TICAH 2011). Despite situational differences in materials and methods between countries and regions, it appears that there are common cultural reasons behind the use of traditional additives, such as flavouring and to hasten cooking (Kaputo 1996; Wanjekeche *et al.* 2003).

2.4. Source of study materials

Materials obtained in Uganda are described here, as they were used for all study objectives and as such, apply to Chapters 4, 5, and 6. Legumes are a common staple food in Uganda (Rubaihayo 1995; Minka, Mbofung, Gandon, & Bruneteau, 1999). Black beans (*Phaseolus vulgaris L.*) were chosen for use in this study as it is a legume which is both popular and regularly prepared with plant ash filtrate (A. Akello, NUDF Women's Group, pers. comm. 2012). Dried black beans were purchased in bulk in Lira, Northern Uganda from a vendor at the Lira Central Market. The origin and storage history of the beans was unclear; however it is known that they were harvested during the 2011 season in the Lira area, dried, and stored without preservative. Approximately 1 kilogram (kg) of dried beans were sealed in a plastic Ziploc™ bag for transport to Canada while the remainder was used for in-country preliminary cooking trials and the palatability tests. Two 500 gram (g) bags of ground salt were purchased from vendors at the Lira Market, and both came from the Lake Magadi area in Kenya. One 200 g sample was drawn from each supply, sealed in 250 ml wide mouth high-density polyethylene (HDPE) containers, and labeled with description, date, and geographical coordinates. These samples were stored dry and without preservatives for later analysis in Canada. A portion of the remainder was used in Uganda for preliminary cooking time trials and for cooking beans for palatability tests. One 500 g bag of Chiluma iodized commercial sea salt, originating from Kensalt Limited in Kenya, was purchased in Kampala. One 200 g sample was drawn, sealed in a 250 ml wide mouth HDPE container, and labeled with description, date, and geographical coordinates. This sample was stored dry and without preservative for analysis in Canada, while the remainder was used in Uganda for preliminary cooking time trials and for cooking beans for palatability tests.

2.4.1. Explanation and procurement of plant ash filtrate

Plant ash is obtained in rural Uganda from the remains of specific harvested crops. During the field data collection in June 2012, traditional methods of plant ash collection and production were observed and recorded. The crop most often used for making filtrate is legumes, which includes several varieties of the common bean (*P. vulgaris*); soy, black, kidney, and yellow, as well as peanuts (*Arachis hypogaea*). The other plant identified as most commonly used for ash filtrate was sesame (*Sesamum indicum*). These crops are typically harvested once per year, usually in conjunction with the shorter dry season from June to early August. Once the crops are mature, whole stalks (including leaves, empty pods, and roots) are harvested and bundled for transport to a drying area, which is typically a large swept dirt space near the farmer's home. Plants are spread out during the day and piled under cover at night until thoroughly dry (Figure 3). When ready, stalks are manually thrashed using a stick or similar instrument to remove the beans or seeds from the pods (Figure 4).

Figure 3. Curing of a harvested *Phaseolus vulgaris* L. crop.

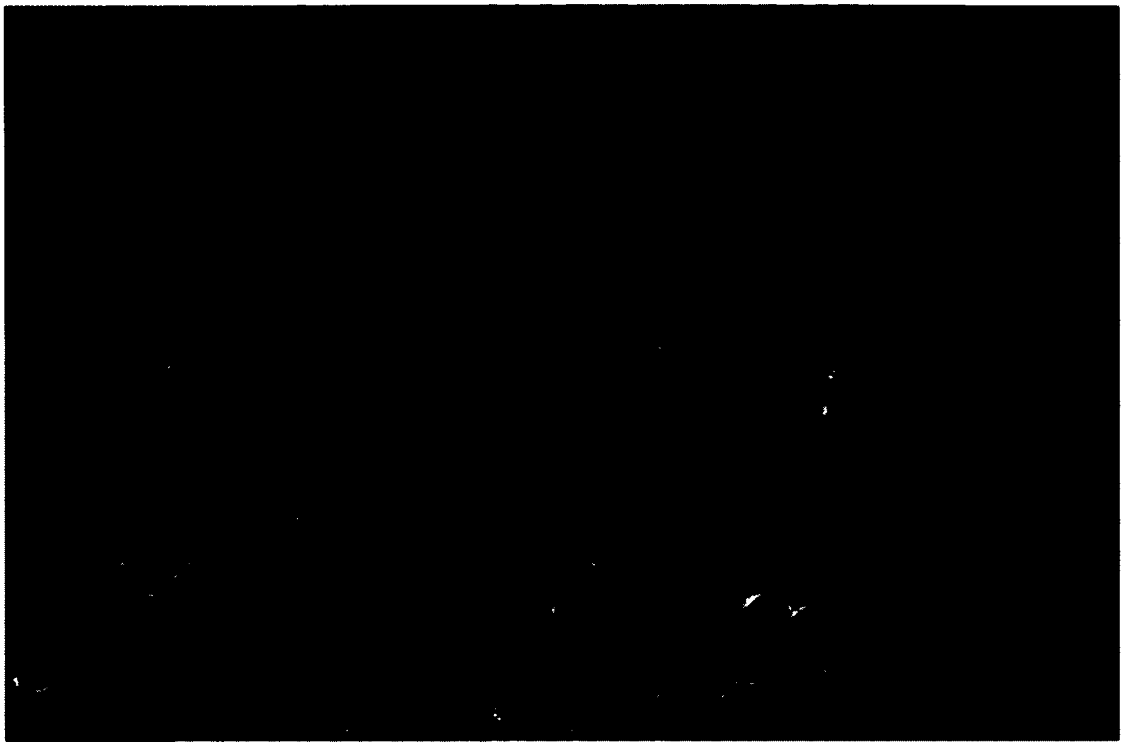

Figure 4. Demonstration of manual thrashing of a dried legume (*Phaseolus vulgaris* L.) crop.

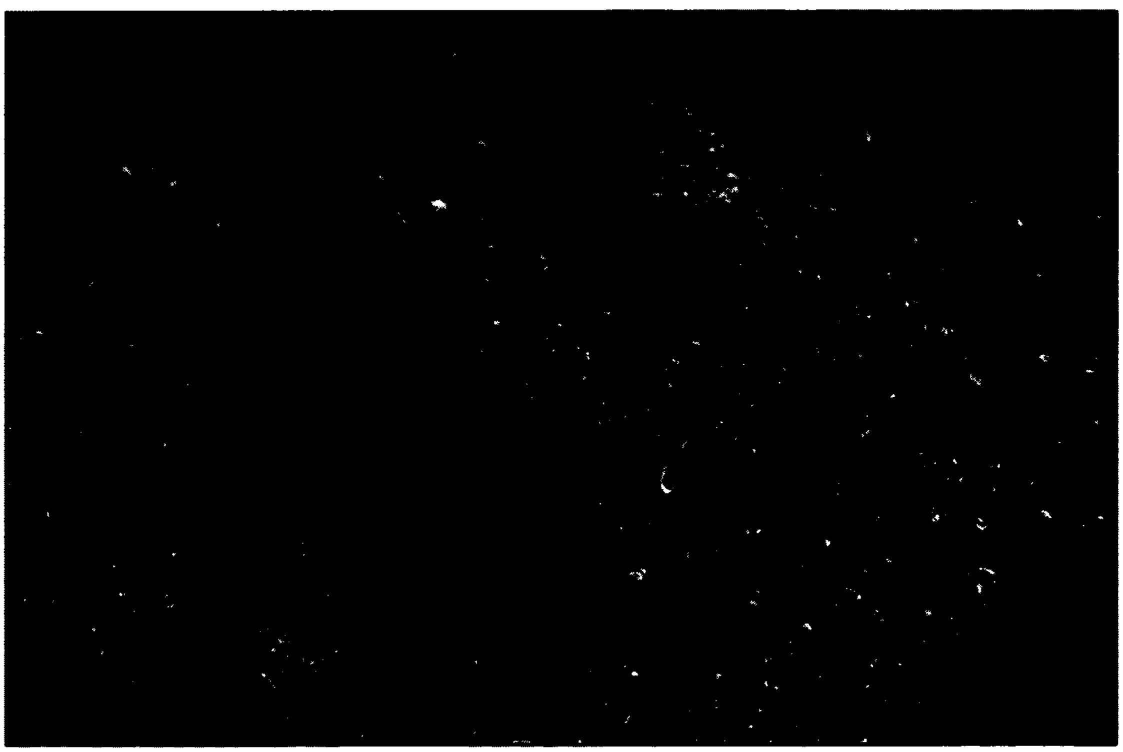

Figure 5. *Phaseolus vulgaris* L. crop residue ash ready for use.

The stalks are then separated from the beans or seeds and placed in another swept dirt area in large piles for burning. Once cooled, the ash (Figure 5) is scooped from the ground into pots or bags for storage to be used throughout the year. Banana peels (*Musa acuminata*) was also mentioned as a possible ash source, although it appears to be used less frequently in this region.

Ash filtrate is made as required for cooking, and excess will sometimes be kept for subsequent days' use. From storage, approximately 250 g of loose, dry ash is put into a small sieved container on top of a rudimentary filter. The filter is made from a local spear grass which has been ripped into short lengths and placed in the bottom of the container (Figure 6).

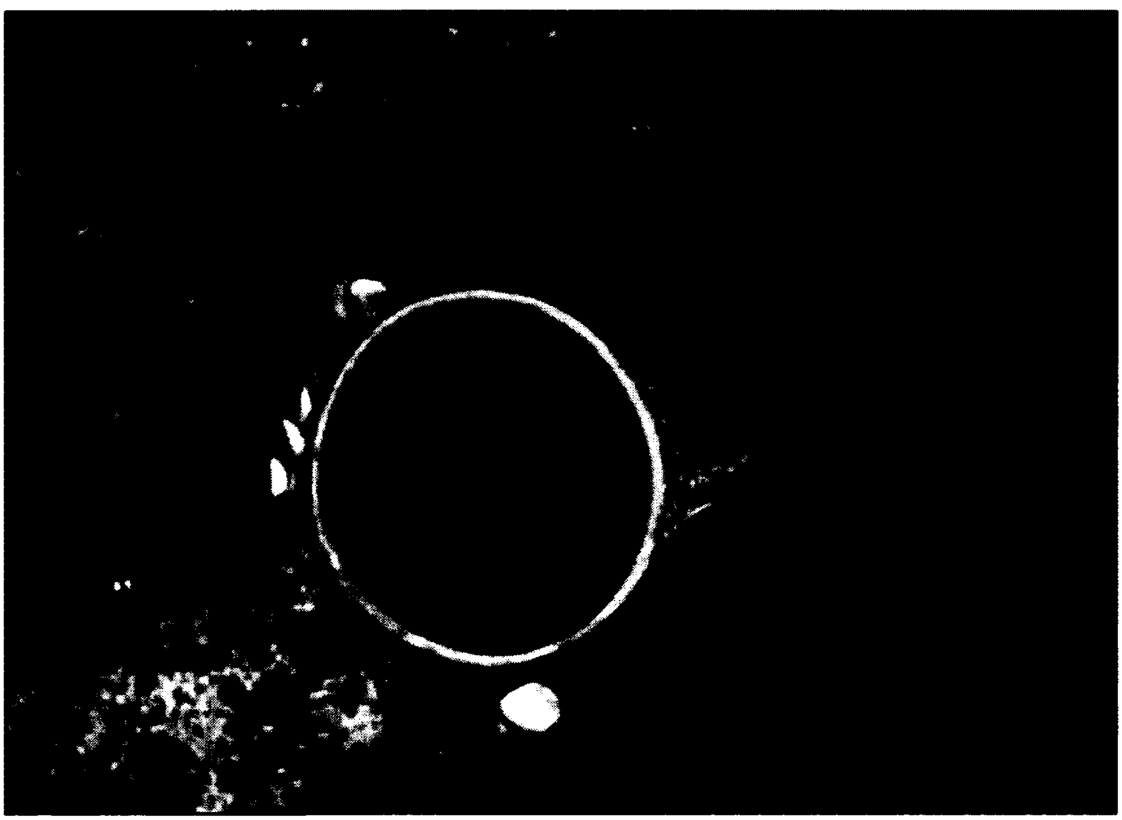

Figure 6. Traditional spear grass filter used for making ash filtrate.

Water is slowly added to the ash until it is just saturated, and then the ash is pressed firmly down. More water is then added to fill the top container, which slowly percolates for approximately 20 minutes. As it drops, the filtrate is collected in a receiving container (Figure 7). This filtrate is then added by the spoonful directly to cooking water.

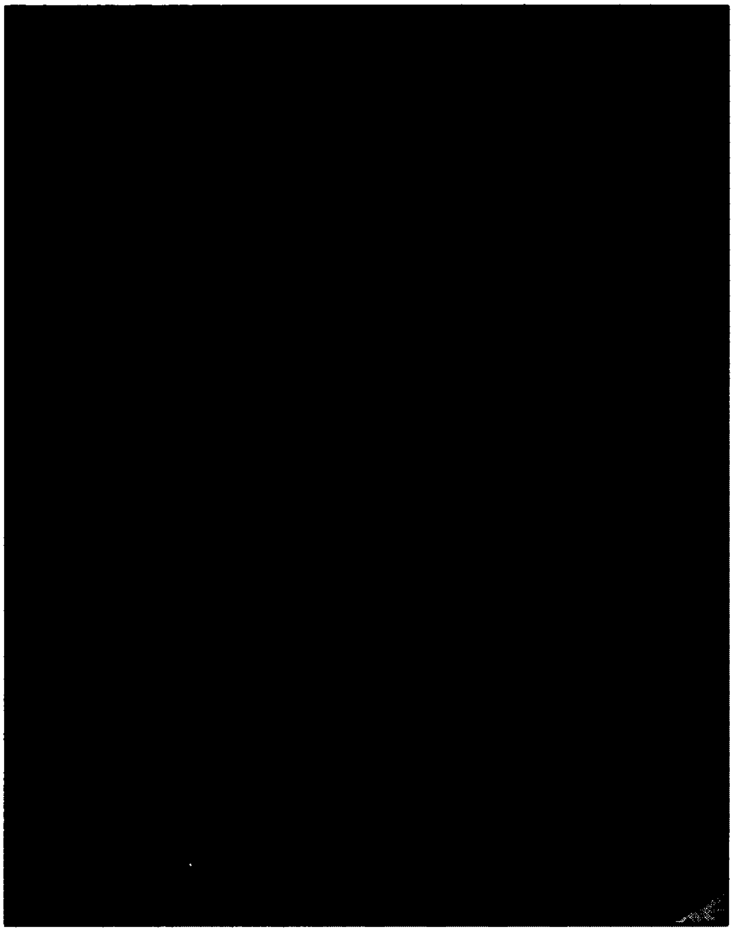

Figure 7. Pressed plant ash on filtering bed (upper cup) and subsequent filtrate after percolation (lower cup).

Two 150 g plant ash samples were collected from each of two households at each of the four study sites from interviewees (n=2x2x4=16). Ash collected represented both the 2011 and 2012 harvest years. While the intent was to collect individual random samples, lack of census data about households and access issues prevented this. However, it was observed that harvest and ash burning are community activities, and the ash is shared among several families within villages. Ash was collected from thoroughly mixed homogenous supplies at each study site, to ensure representative samples. The ash was dry at time of sampling, collected with a plastic scoop, and filled 250 ml wide mouth HDPE containers. Each container was sealed and labeled with date, study site, sample identification number, year of procurement, and geographical coordinates.

Chapter 3. Literature Review - Salt additives used for cooking legumes

3.1. Commercial salt

The use of monovalent salts, like commercial table salt and sodium bicarbonate, as cooking additives has been extensively documented, especially with regards to cooking time and improving flavour. These types of salt, particularly table salt, are the most widely known and abundantly used throughout the world. Salt can have noticeable effects when used for the preparation of hard-to-cook foods like legumes, in the softening of texture (Silva, Bates, & Deng, 1981; Van Buren 1986) and in reducing the length of required cooking time (de León *et al.* 1992; Onwuka & Okala 2003). Plant cell walls contain pectins which play a key role in maintaining structural integrity of the cells. The addition of monovalent salts to cooking water leads to degradation of the pectic substances, among other processes, which in turn causes the legume to soften (Van Buren 1986; Liu, Philips, & McWatter, 1993).

The decrease in cooking time has been suggested to be a result of several processes, including pectin solubilization (Van Buren 1986; Uzogara *et al.* 1990), ionic interchange and chelation (Varriano-Marston & de Omana 1979; de León *et al.* 1992), improved heat transfer properties (Coyoy Gonzalez 1987), increased water absorption capacity (Varriano-Marston & de Omana 1979), and increased water holding capacity (Garcia-Vela & Stanley 1989). At a concentration of 0.6 g/L, NaCl reduced the necessary cooking time from 55 minutes (control) to 44.5 minutes for cowpeas (*Vigna unguiculata*), while the $NaHCO_3$ sample took only 38.5 minutes (Onwuka & Okala 2003). The use of divalent salts like calcium chloride ($CaCl_2$) caused an opposite reaction, where legumes actually took 50 minutes longer to cook than the control sample. Ionic interchange is described by de León *et al.* (1992), who studied the effect of salt solutions on several characteristics of common beans by varying the ratio of monovalent (Na^+ and K^+) to divalent (Ca^{2+} and Mg^{2+}) ions. Cooking time for dried beans decreased significantly by increasing the ratio of monovalent to divalent ions, for beans presoaked in the solution and cooked in water, from over 6 hours (control, 4.6:1) to approximately one half hour (9.8:1) (de León *et al.* 1992). These results were replicated by Onwuka & Okala (2003), where different concentrations of various salts were analyzed for impact on cooking times of two legumes. For both legumes, the addition of NaCl or $NaHCO_3$ significantly reduced cooking time in comparison to the control, while the addition of the divalent salt calcium chloride ($CaCl_2$) increased cooking time.

The other main reason for cooking with table salt is to improve palatability, both in legumes and many other foods. To test the palatability of legumes, several properties are often examined including texture, flavour (taste), colour, and overall acceptance (Silva *et al.* 1981; Onwuka & Okala 2003; Wanjekeche *et al.* 2003). Onwuka & Okala (2003) found that the addition of table salt to cooking water scored significantly better than any of plain water, ground salt, sodium bicarbonate, or calcium chloride for all sensory properties. Though it decreases cooking time and improves flavour, there is harm in excessive use. The main issue is that it contains a high amount of sodium, which is well known to contribute to hypertension, a precursor to several health concerns including cardiovascular disease, stroke, and edema (SACN 2003).

3.2. Indigenous salt

Indigenous (ground) salt has been studied mainly for its use in dietary applications and on its mineralogical composition. The term 'indigenous' salt typically refers to an unrefined and impure salt. In Northern Uganda, the indigenous salt used is a precipitate from large saline lakes located in Kenya and is referred to as 'magadi'. The name varies with location however, and so in this thesis it will be called 'ground' salt. Mineralogical, chemical, and physical properties of ground salt differ slightly among studies; however, it is generally acknowledged that all contain the mineral 'trona', a hydrated sesquicarbonate of sodium, making it highly alkaline (Makanjuola & Beetlestone 1975; Nielsen & Dahi 2002; Mamiro *et al.* 2011).

Similar to commercial salt, ground salt is added to cooking water in Africa to both decrease cooking times and improve flavour. The alkaline nature of ground salt acts to tenderize hard-to-cook foods, like legumes, by improving cell membrane permeability (Wanjekeche *et al.* 2003), pectin solubilization, starch gelatinization, and cell swelling (Uzogara *et al.* 1990). The addition of ground salt to cooking water decreases required cooking time for dried legumes to varying degrees, depending on the concentration. Ankrah & Dovlo (1978) found a reduction in cooking time of 20-35 minutes for cowpeas at a concentration of 0.05 g of ground salt to 25 g legumes in 500 ml of water. Similarly, Onwuka & Okala (2003) showed a decrease in required cooking time of 13 minutes for cowpeas and 24 minutes for African yam beans (*Sphenostylis sternocarpa*) at a concentration of 0.6 g/L. Most recently, it was found that cooking time for beans could be reduced by as much as 60 minutes with the addition of 20 g of ground salt to water for 0.5

kg of beans (Mamiro *et al.* 2011). It appears that increasing the concentration of indigenous salt has a substantial effect on decreasing cooking time.

Despite the benefits, there are disadvantages to adding a high amount of ground salt to cooking water. Beans turned dark or brown, making them less visually desirable (Ankrah & Dovlo 1978; Onwuka & Okala 2003), especially compared to beans cooked in plain water (Wanjekeche *et al.* 2003). The distinctive flavour imparted by the ground salt seems to elicit mixed opinions, in some areas (e.g., Tanzania) it is apparently liked (Mamiro *et al.* 2011) while in others (e.g., Nigeria) it is not (Onwuka & Okala 2003). Further, the high alkalinity of this cooking water could have a destructive effect on the mineral bioavailability of certain elements (Mamiro *et al.* 2011) and cause a significant decrease in levels of essential amino acids (Minka *et al.* 1999).

The erratic amount of various minerals found in different indigenous salt samples has also prompted questions about the health effects of its use (Sodipo 1993). In addition to high sodium, elevated levels of iron, fluoride, and manganese have been found (Ankrah & Dovlo 1978; Nielsen & Dahi 2002; Mamiro *et al.* 2011). High fluoride exposure from use of ground salt has been linked to severe dental fluorosis (pitting of teeth) in Tanzania (Mabelya, van Palenstein Helderman, van't Hof, & Konig, 1997), while excess iron exposure can contribute to iron overload (Gordeuk *et al.* 1992).

3.3. Plant ash and filtrate

The practice of using dried plant ash or for the creation of ash filtrate as a type of salt has a long history. These practices have been documented, though sporadically, in Africa and several other parts of the world. In the mid-1900s, Culwick (1950) noted the procurement of minerals and salts from plants in a book about the diets of the Azande in Sudan. The traditional practice described in this book is very similar to the current practice of using crop ash filtrate, although once a filtrate was made it was allowed to evaporate, and the residual salts were then used (Culwick 1950). Around the same time, a practice was observed in Western Kenya, where filtrate was added to various foods for flavouring (Huntingford 1955). Work in the 1970s capitalized on advancing technology and analyzed the components in ash made from the sago palm tree (*Metroxylon* spp.) in Papua New Guinea (Townsend *et al.* 1973). In this setting, the dry ash was eaten alone on an irregular basis, during celebratory rituals and on an opportunistic basis when it was available. The alkaline sago ash likely supplemented certain mineral requirements in the predominantly vegetarian diets, as a typical 2 g serving would provide 12 mg sodium, 526 mg potassium, 234 mg calcium, and 69 mg magnesium (Townsend *et al.* 1973). The Hopi tribes of the United States traditionally added ash from residual bean pods, four-wing salt bush (*Atriplex canescens*), and other plants to corn foods, probably to augment mineral content (Calloway *et al.* 1974; Kuhnlein *et al.* 1979). The presence of lead, a toxic metal was discovered in the plant ash from this site, in addition to high levels of strontium, rubidium, and copper, each of which may pose health risks if ingested in sufficient quantity (Kuhnlein 1980). Plant ash filtrate in Zambia was also found to be highly alkaline, and contain elevated levels of sodium, potassium, calcium, iron, and copper (Kaputo 1996). Women in rural areas reportedly still use the filtrate from groundnut (peanut) shells, banana leaves, and bean stalks quite often in preparation of a traditional meatless dish (although sodium bicarbonate was sometimes being chosen instead, suggesting its suitability as a replacement). Wanjekeche *et al.* (2003) were probably the first to study the effect of plant ash on cooking time, with comparative studies on bean ash (unidentified species), maize cob ash, ground salt, citric acid, and plain water. Both the ash treatments as well as the ground salt treatment were effective in reducing cooking time, but had negative effects on the nutritional content and on the panelists' acceptability of colour and taste of the cooked product. In the most recent and comprehensive study to date, Mamiro *et al.* (2011) compared functional and nutritional effects of using ground salt and plant ash to cook several foods. Despite a wide variation in levels of elements, the addition of either ground salt or plant ash to cooking water significantly reduced cooking time for all foods compared to tap water. Both additives were also found to be of high alkalinity which decreased the bioavailability of certain minerals, including zinc and iron.

Due to the obscurity of this practice, there are two fundamental gaps in knowledge which require understanding: first, whether the anecdotal beliefs about the use of this filtrate are valid (i.e., Does the use of ash filtrate actually decrease cooking time and make food more palatable?); and second, whether the regular use of ash filtrate by individuals could have dietary effects of potential health concern.

3.4. Thesis objectives

The intent of this research is to enhance local food security and human and environmental health through an examination of traditional practices related to using ash filtrate in food preparation in Northern Uganda. Four objectives will contribute to the understanding of this practice: 1) compare cooking times for a common Ugandan food (*Phaseolus vulgaris* L.) in plain water (control) and water containing known concentrations of commercial salt, local ground salt, and ash filtrate (treatments); 2) assess palatability of the common food cooked in each treatment; 3) assess whether specific socio-economic factors affect palatability preferences and use of the cooking practice; and 4) determine the chemical composition of the ash and ash filtrate to assess and identify potentially toxic or unhealthy constituents. The information from this work will help outline education and policy recommendations about this practice and provide a platform from which to base future studies about similar unknown practices. Given the known extent of this practice in Africa, it is imperative to understand what elements are being consumed and their consequent effects on human health.

Chapter 4. Crop ash filtrate influence on cooking time of dried black beans (*Phaseolus vulgaris* L.)

4.1. Abstract

Filtrate made from the burnt ash of crop plant residue is a traditional additive used in cooking in Northern Uganda. It is believed by locals that this additive decreases the cooking time of hard- to- cook legumes. The intent of this study was to compare cooking times for dried black beans (*Phaseolus vulgaris* L.) among four treatments: Type 1 water (control), table salt, crude ground salt, and ash filtrate. Analysis of variance (ANOVA) with post-hoc Bonferroni multiple comparisons showed a statistically significant ($p < 0.01$) 27% reduction in cooking time with the addition of ground salt and 18% shorter cook time with ash filtrate, substantiating the anecdotal belief held by locals. Commercial salt slightly increased the cooking time compared to the control. The reduction in cooking time has important implications for fuel wood requirements as the majority of households rely on fires for cooking. However, there is uncertainty about the health effects of using plant ash filtrate requiring further investigation. As an alternative to the use of plant ash filtrate, presoaking of dried legumes is suggested as a means to decrease cooking time while precluding any potential health issues associated with the consumption of ash filtrate. Further research in these communities is necessary to determine both the acceptability of incorporating presoaking beans into food preparation practices and what educational opportunities may be the most effective.

4.2. Introduction

Legumes are one of the best sources of vegetable protein available in developing countries (Siegel & Fawcett 1976) and are a staple food in Uganda (Mamiro *et al.* 2011). Black beans (*P. vulgaris*) are particularly common in Northern Uganda, as both a domestic crop and commercial product in markets. The typical climate of Uganda, humid and moderately hot (20-30 °celcius (C)), creates poor storage conditions for legumes (Reyes-Moreno & Paredes-Lopez 1993). Drying and storage in these conditions result in beans becoming hard-to-cook; a property which requires extensive cooking time to make them palatable, eliminate toxic components, and allow for the protein to become nutritionally available (Varriano-Marston & de Omana 1979; de León *et al.* 1992). Salt is regularly used to decrease the cooking time of dried, hard-to-cook legumes (Uzogara *et al.* 1990; Onwuka & Okala 2003; Mamiro *et al.* 2011). Depending on availability, people of different regions use several different types of salt or salt-like additives for cooking. These may include refined commercial (table) salt (NaCl), a crude indigenous (ground) salt precipitated from saline lakes, and/or the ash of plant parts.

The cooking of legumes involves several processes to render them palatable, digestible, and increase nutrient availability. A main component of plant cell wall structure includes pectic substances, which need to be degraded in order for internal cell tissue, and therefore the bean, to soften (Uzogara *et al.* 1990). Commercial (table) salt decreases cooking time for legumes by accelerating the degradation of this pectin (Van Buren 1986), through ion exchange and chelation where ions of the 'intercellular cement' are either replaced by sodium ions or leached out (Varriano-Marston & de Omana 1979). In some parts of the world, an indigenous ground salt is also used to speed up cooking time of legumes. Ground salt is composed mainly of trona, a hydrated sesquicarbonate of sodium, which makes it highly alkaline (Makanjuola & Beetlestone 1975). Alkalinity has been shown to have great influence on the softening of legumes, probably due to an effect on the starch gelatinization process (Wanjekeche *et al.* 2003) and in turn, decreases the necessary cooking time (Ankrah & Dovlo 1978; Uzogara *et al.* 1990; Onwuka & Okala 2003). As an alternative to ground salt, plant ash and/or plant ash filtrate is used as a traditional cooking additive in some areas of the world, including rural Africa (Kaputo 1996; Wanjekeche *et al.* 2003; Mamiro *et al.* 2011). Plant ash and ground salt are both highly alkaline, and have been shown to have elevated mineral content; both factors which probably contribute to the observed reduced cooking time. Previous research has found no significant difference in the cooking time of beans when either plant ash or ground salt have been added (Mamiro *et al.* 2011).

In rural Northern Uganda, there is anecdotal information that a filtrate made from residual crop ash also considerably reduces the length of time required to cook dried legumes. The plants most often burnt for ash are crop residues of legumes, and sesame (*Sesamum indicum*) (Chapter 2.4). While there is ample information on the use of sodium salts to hasten the cooking of legumes, research on the use of crop ash and filtrate as an additive is limited. Further, there have been no simultaneous direct comparisons of the effect of all three types of salt additives on the cooking time of legumes. The objective of this study was to compare the time required to cook dried black beans in four treatments:

Type 1[1] water (control), ground salt, table salt and ash filtrate. It is hypothesized that all treatments will decrease cooking time compared to the control.

4.3. Materials and methods

4.3.1. Study site

Controlled cooking time trials took place in the Central Equipment Laboratory at the University of Northern British Columbia (UNBC) in Prince George, British Columbia, Canada (10U 512250m E, 5971618m N) at an elevation of 775 metres above sea level. Field interviews were conducted in Oyam and Lira districts, Northern Uganda.

4.3.2. Treatments and sample preparation

A preliminary cooking time trial took place in Kampala, Uganda to gain understanding about the use of traditional additives (e.g., concentration), the amount of water required, and the time requirements for conducting this study. Lack of electricity prevented the trials from being carried out closer to the study sites, however guidance about the use of ground salt and filtrate was provided by a local woman in Kampala. The information gained during these trials was used to conduct the controlled cooking trials at UNBC.

The study design consisted of four cooking treatments for black beans (*Phaseolus vulgaris* L.) (Type 1 water – 'control', water containing commercial salt – 'table salt', water containing local ground salt – 'ground salt', and water containing plant ash filtrate – 'filtrate'). Dried black beans were sorted by hand, and shriveled beans and other detritus were removed. Four 2.3 litre (L) Starfrit Starbasix® saucepans were thoroughly washed

[1] Ultrapure water filtered with a Millipore® system meeting ASTM® standards for Type 1 water.

three times prior to use, rinsed with Type 1 water, and dried. They were then prepared with 80 grams (g) of dried beans, 1.0 L of Type 1 water, and one of the four treatments. The amount of water required for cooking black beans was based on the preliminary trials done in Kampala and at UNBC, and the treatment concentration was determined through interview responses. For 80 g of dried beans, the following amounts were used: 2 g of commercial salt, 2 g of ground salt, and 15 ml of ash filtrate. These concentrations of table salt (0.2% w/v), ground salt (0.2% w/v), and ash filtrate (1.5% v/v) are comparable with similar studies (Onayemi, Osibogun, & Obembe, 1986; Wanjekeche *et al.* 2003). For a full description of materials and additives, please refer to Chapter 2.4. The ash filtrate was prepared using a method as similar as possible to that done in Uganda. Approximately 250 g of loose, dry ash was placed into an unbleached coffee filter. The filter was used in place of the rudimentary spear grass filter because such grass was not available. The filter with ash was placed into a perforated (0.5 cm holes at the bottom) plastic cup from Uganda. A small portion of 200 ml of Type 1 water was added to the ash which was slightly tamped down to form a filter bed. The remainder of the water was slowly added and percolated through, collecting in another plastic cup for use. The Dog Abam ash sample was used for preparing filtrate for this portion of the study.

4.3.3. Study design

The beans were cooked individually on four separate Toastess® cooking ranges (THP432). Each cooking range was preheated to maximum temperature (550°C), as indicated by the automatic shut off switch, and covered pots were placed simultaneously at the maximum temperature. The pH of cooking water of each treatment was taken using a

Bluelab® pH pen immediately after mixing in the respective additive to the cooking water; just prior to placing pots on burners. Values of pH were recorded for each treatment once the reading held steady for 10 seconds, and the pH pen was rinsed in Type 1 water prior to subsequent test. Temperatures of prepared pots were identical at the time of placement, and were recorded at time of boiling and time of completion using individual UEi™ DT15A Digital Thermometers. Beans were considered cooked and the time recorded when puncture force registered as 150 g (Silva *et al.* 1981), as measured with a Mecmesin gram gauge DGD-6. Beans continued cooking until considered soft by hand and mouth feel (Ankrah & Dovlo 1978; Onwuka & Okala 2003; Wanjekeche *et al.* 2003); where beans easily ruptured between fingers and had attained a consistent softness, and time was recorded again. This was done for comparative purposes, as the consistency of beans considered cooked at 150 g of puncture force was still chalky, and not palatable according to preference standards observed in Uganda. This cooking time trial procedure was repeated three times at UNBC for replication purposes.

4.3.4. Informal field interviews

Semi-structured interviews (see Appendix A for list of questions) were conducted to gain local perspective and a cultural context, adding to a more comprehensive understanding of this practice. Due to a lack of census data, interviews were not able to be conducted randomly. Instead, a local research assistant/translator (Ugandan female, aged 40) who was hired to assist with field work acted as a key informant, providing introductions to women representing a variety of demographics. Prior to data collection, a meeting was held with the local research assistant to explain the process and goals of this study, answer any questions, and ensure privacy for all participants through a signed confidentiality agreement (Appendix B). I conducted two trial interviews to clarify and assess appropriateness of questions, determine length of interviews and gain familiarity in working with the research assistant. All interviews ($n = 20$) were conducted via translation at the homes of each respondent and were recorded on paper.

4.3.5. Statistical analysis

Data was tested for normality using the skewness and kurtosis test, and all variables met the assumptions of normality. Homogeneity of variance was confirmed by Levene's statistic. Post-hoc Bonfferoni multiple comparison tests were used to determine significant differences among treatment means (Gotelli & Ellison 2004). The conservative nature of using a Bonfferoni test ensured significantly different cooking times as applicable to actual situations where at least a moderate time difference would be required to have noticeable implications for cooking time. Statistical significance was determined with $\alpha = 0.05$ and the null hypothesis was that no difference exists among means between control and treatments.

Analysis of variance (ANOVA) tests were used to assess significant differences among means of response variables associated with cooking treatment. The mathematical model used was:

$$Y_i = \mu + treatment_i + \varepsilon_i$$

where Y_i = response variable mean (pH, temperature of cooking water treatment at boiling, temperature of cooking water treatment at 15 minutes, cooking time to 150 g puncture force test, or cooking time to palatable); μ = grand mean response variable; $treatment_i$ =

cooking water treatment (Type 1 water [control], commercial salt, ground salt, ash filtrate); and ε_i = experimental error.

Statistical analysis was done using Stata® (Version 12). Compilation of interview responses and determination of percent responses were completed using Microsoft® Excel 2010.

4.4. Results

4.4.1. Experimental evidence and cooking times among treatments

Differences among treatment solutions are presented in Table 2. Cooking water pH levels were significantly ($p < 0.05$) different among all treatments, with ground salt (9.6 ± 0.06) and ash filtrate (10.5 ± 0.03) being the most alkaline. The temperature of cooking water, both at time of boiling and the average throughout cooking, did not differ significantly ($p > 0.05$) among treatments. However, despite similar temperature, a statistically significant difference among means was found for the time to boil but only between the control (587 ± 3.33 seconds) and ground salt treatments (502 ± 15.95 seconds). A significant difference among means was found for cooking time necessary to obtain a puncture force of 150 g. The addition of ground salt significantly ($p \leq 0.01$) decreased time by up to 18% and ash filtrate by up to 13%, compared to either the control or table salt treatments. The required cooking time to acceptable texture increased for all treatments except ground salt, which felt palatable and not at all chalky when the puncture force test was passed. A significant ($p \leq 0.05$) difference among treatment means was found for cooking time to acceptable texture. The ground salt and ash filtrate treatments decreased cooking time to eating soft significantly ($p \leq 0.01$), compared to the control, by 30.3 and 20.3 minutes or 27% and 18%, respectively.

Table 2. Mean (± standard error) treatment values of pH, temperature, time to boil and cooking times ($n = 3$). Values along rows with the same superscript are not significantly different at $p \geq 0.01$ by the Bonferroni method.

	Treatment			
	Control	**Table Salt**	**Ground Salt**	**Ash Filtrate**
pH of cooking treatment	$7.7^{*} \pm 0.07^{a}$	8.2 ± 0.03^{b}	9.6 ± 0.06^{c}	10.5 ± 0.03^{d}
Temperature at boiling (°C)	97.6 ± 0.19^{a}	97.5 ± 0.13^{a}	97.6 ± 0.13^{a}	97.6 ± 0.09^{a}
Temperature at 15 minutes (°C)	97.8 ± 0.07^{a}	97.9 ± 0.06^{a}	97.7 ± 0.09^{a}	97.6 ± 0.07^{a}
Time to boil (seconds)	587.0 ± 3.33^{a}	553.0 ± 19.47^{ab}	502.0 ± 15.95^{b}	562 ± 17.24^{ab}
Cooking time to 150 g puncture force (minutes)	101.0 ± 0.88^{a}	101.3 ± 0.88^{a}	$82.7^{**} \pm 0.88^{b}$	88.3 ± 0.67^{c}
Cooking time to eating soft (minutes)	113.0 ± 1.15^{a}	121.7 ± 1.20^{b}	$82.7^{**} \pm 0.88^{c}$	92.7 ± 1.20^{d}

*pH analysis of pure Type 1 water may not yield accurate results due to lack of elements and conductivity in the water.
**The texture of beans cooked in the ground salt treatment was deemed satisfactory according to Ugandan preferences at the time of puncture force test, demonstrated by the same value for both of the cooking times. This texture was used as a comparative threshold for other treatments.

4.4.2. Participant responses relating to cooking times and treatments

Statistical findings supported interview responses, where all women stated that ash filtrate decreased cooking time and that the difference could be 2-4 hours faster depending on the amount being cooked and the size of the fire (Figure 8). Other responses about actual time savings were less definitive but still suggested considerable time savings; "There is a big difference between food cooked with *kado atwona* [ash filtrate]...it is much faster". Several respondents did say that ground salt could be used instead of ash filtrate, both for decreasing cooking time and to make the taste culturally acceptable. All interviewees further specified that substituting table salt for the ash filtrate was not possible because it wouldn't have the same effect on cooking time. However, several women said that they use both ash filtrate to speed cooking and table salt after cooking to improve flavour.

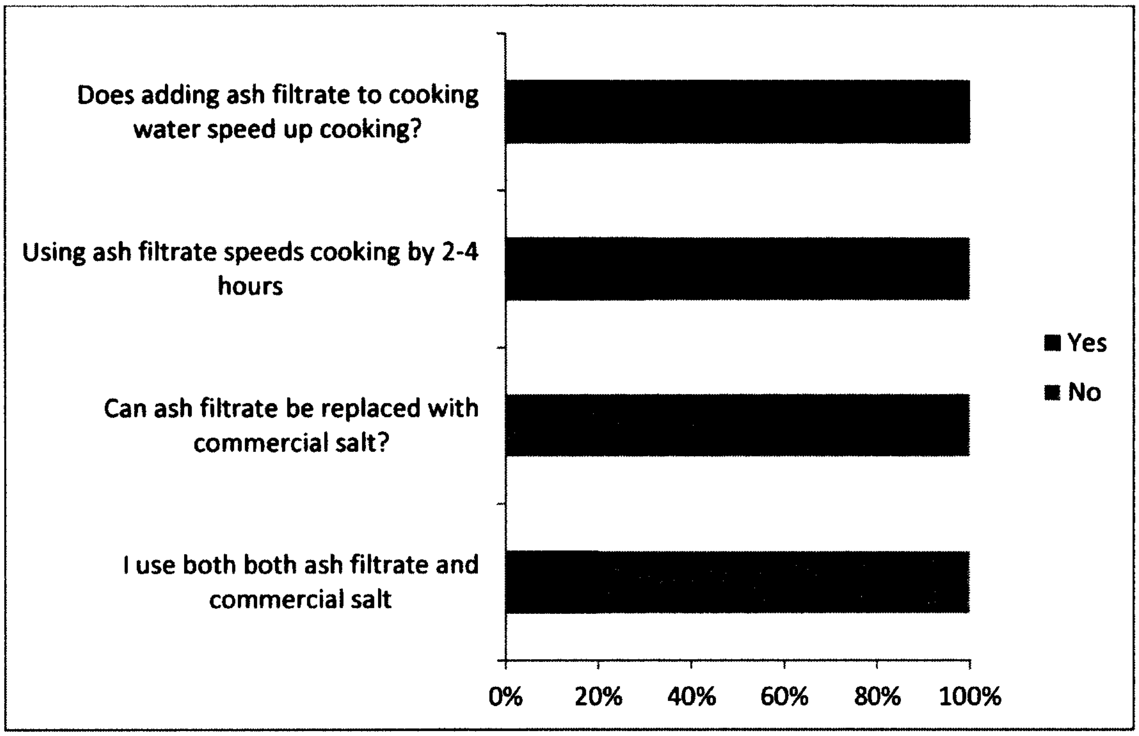

Figure 8. Percent response of cooking time related interview responses (n=20).

4.5. Discussion

Ash filtrate decreased cooking time as compared to either the control or table salt treatments, validating the anecdotal belief. Additionally, ground salt was found to decrease cooking time even more than the ash filtrate. Based on this finding that both traditional treatments sped up the cooking process, it is probable that they share similar properties and/or mechanisms of pectin degradation, confirming previous findings (Mamiro *et al.* 2011). Indeed, both treatments were highly alkaline (pH 9.6 (ground salt) and 10.5 (ash filtrate)) compared to either the control or table salt treatments (pH 7.7-8.2, respectively), and both contained high levels of elemental concentration, particularly sodium (ground salt) and potassium (ash filtrate) (Chapter 6.4.1.). The slightly basic pH of the table salt treatment is likely due to additional impurities in the salt (Varriano-Marston & de Omana 1979). High concentrations of sodium and potassium, and an alkaline pH have all been shown to be important in the reduction of cooking time of legumes (Varriano-Marston & de Omana 1979).

As the temperature at boiling was not found to be significantly different among treatments in this study, it could not be the cause of differing cooking times. The addition of a solute (e.g., salt) to a pure solvent (e.g., water) is known to elevate boiling temperature due to vapor pressure differential between the pure solvent and the solution (Andrews 1976). This colligative property depends on the number of particles present in the solution. The lack of observed difference in boiling temperature in this study is likely because a small amount of solute was added, which did not contribute sufficient particles to change the temperature at a scale observable with our thermometers.

Another interesting anomaly, with respect to many other studies, was that the table salt treatment did not decrease bean cooking time as compared to the control, and actually took longer than the control to become texturally acceptable (Table 2). This result was found in both the preliminary trials in Uganda and the controlled cooking trials done at UNBC, with similar cooking equipment and identical test materials used for both. The use of NaCl has consistently been shown to increase the rate of softening and therefore decrease cooking time for hard- to- cook beans (Silva *et al.* 1981; Van Buren 1986; de León *et al.* 1992). de León *et al.* (1992) found continually significant decreases in cooking time obtained with increasing ratios of monovalent to divalent ion concentration (increased Na^+, predominantly). By the methodology explained in de León *et al.* (1992), the ratio of monovalent to divalent ions in the plain (control) oven-dried beans (80 g) in this study was 4.2; the sum of monovalent ions (K^+ and Na^+) was 964 mg (919 mg K^+ + 45 mg Na^+) and the sum of divalent ions (Ca^{2+} and Mg^{2+}) was 228 (104 mg Ca^{2+} + 124 mg Mg^{2+}) (see Chapter 6.4, Table 9). The ratio of 4.2 in this study was lower than the 4.6 ratio of the dried beans used in de León *et al.* (1992). The addition of 2 g of table salt, 2 g of ground salt, and 15 ml of ash filtrate treatment additives to 1 litre of water and 80 g of dried beans would alter the ratio to 7.4, 4.9, and 5.1, respectively (see Chapter 6.4, Tables 5 and 8). In contrast to previous findings, the addition of table salt and increased monovalent to divalent ratio in this study did not decrease cooking time compared to the control. This may be due to differences in chemical composition of the commercial salts used in each study as refining processes are likely to differ between companies and countries of origin. The commercial salt used in this study could contain amounts of additional divalent ions not present in the solutions employed by de León *et al.* (1992). The different outcome may also be due to differences in methodology; de León *et al.* (1992) soaked the dry beans prior to cooking and in this study beans were cooked from the dried state.

A major benefit to the shortened cooking time is the lessened pressure on fuel wood supplies. Deforestation continues to be a problem in Uganda, where the vast majority of the rural population uses wood or wood products (e.g., charcoal) for all domestic energy needs. It has been estimated that the average rural western Ugandan family will use approximately 8.4 kg of fuel wood per day for cooking meals (Wallmo 1996), translating into slightly more than 3000 kg per year. Given that beans and legumes are also the most commonly cooked foods at that study site as well (Wallmo 1996) and that they typically require the longest cooking time on the fire, it is reasonable to assume similar fuel wood requirements in rural Northern Uganda. It is not definitively known whether the practice of using ash filtrate to hasten the cooking of beans is also used in western Uganda; however, it has been suggested by a local contact that this is the case (G. Odongo, personal communication, 2012).

Experience gained by the cooking of beans for the palatability study portion of this research and multiple observations of meal preparation provided basic data on the length of time needed to cook beans on both charcoal stoves and over a fire. When the ash filtrate was added, a medium sized (approximately 1.5 L) pot of beans required about two and a half hours to cook on a charcoal stove in a sheltered environment. While charcoal is becoming somewhat more prevalent in certain areas due to its benefits (e.g., long lasting nature, steady heat provision, and decreased emissions when cooking inside a building), many families cannot afford it. Fuel wood remains the dominant resource for cooking and other domestic requirements (van Gemert *et al.* 2013). Through observation and discussion, the same-sized pot of beans cooked with ash filtrate would take approximately four hours on a fire stove in a sheltered area. By extrapolating the findings of the controlled time trials where ash filtrate resulted in an 18% time difference, we could hypothesize that using the filtrate is saving women approximately one hour (63.2 minutes) when cooking over a fire. This translates into a sizable difference in the amount of fuel wood necessary to cook beans either in plain water or with the ash filtrate.

The beneficial aspect of traditional additives in decreasing cooking time must be weighed against probable drawbacks. The high alkalinity of both treatments has been shown to negatively impact the bioavailability of some minerals (Mamiro *et al.* 2011), decrease fibre content (Wanjekeche *et al.* 2003), and have deleterious effects on various vitamins (Kaputo 1996) and amino acids (Minka *et al.* 1999). In contrast, the high concentration of certain elements in the ground salt and ash filtrate may not only counteract the decreased bioavailability, but also be contributing levels of other elements that could be exceeding daily recommended intakes (Chapter 6). Further, several interviewees stated that they also added table salt after cooking in addition to the use of ash filtrate in order to improve flavour. This introduces additional sodium to the consumed product. The amount will vary depending on preference, but based on observation it could range from one half (~3 g) to one (~6 g) teaspoon (730 mg to 1461 mg sodium, respectively, see Chapter 6) in a pot with 4-6 servings of 240 g cooked beans. A diet high in sodium (greater than 1600 mg/day for adults) is known to contribute to hypertension, which subsequently raises health concerns such as cardiovascular disease, stroke, and edema (SACN 2003). Lastly, protein content and availability has been shown to react differently with various types of salt; ground salt may slightly increase protein content (Onwuka & Okala 2003, Wanjekeche *et al.* 2003), while table salt has a negative effect on protein efficiency ratio and protein content (de León *et al.* 1992, Onwuka & Okala 2003).

In light of the uncertainty around using traditional salts, it is useful to consider alternative means of decreasing required cooking time for hard-to-cook foods. Presoaking of legumes is one method which has been proven effective due to the reduction in water uptake time during cooking (Silva *et al.* 1981) and is a feasible alternative in areas with few resources. The degree of success in rehydrating beans relies primarily on two factors: length of time immersed and type of soaking solution. Storage conditions prior to use have also been identified as critical (Molina, de la Fuente, & Bressani, 1975; Silva *et al.* 1981; Sievwright & Shipe 1986); but little opportunity exists for providing optimal storage in rural and impoverished regions. However, the effects of poor storage conditions can be nearly reversed through presoaking measures (Sievwright & Shipe 1986). The soaking time and type of solution have generally been reported as interdependent factors. When plain water is used longer times are necessary and when a salt or alkaline solution is used the time decreases, similar to the results found with cooking due to analogous pectin degradation processes occurring (Varriano-Marston & de Omana 1979). Soaking legumes in plain water for 24 hours decreased cooking time by 50%, while using a 1% potash solution or 4% NaCl solution for 12 hours yielded the same result (Njoku *et al.* 1989). Silva *et al.* (1981) found that after 12 hours no appreciable difference in water uptake could be found, and that the majority of rehydration took place in the first two hours. They suggest that soaking time with plain water should be over eight hours where as little as one hour is needed with a salt solution. Further, there is a more dramatic effect on decreasing cooking time when the soaking solution is retained as the cooking water instead of draining the legumes and cooking with water (de León *et al.* 1992). Shorter soaking times lessen concern over microbial growth, which has been associated with longer soaking times (Hoff & Nelson, 1966).

To date, it does not appear that a study using traditional salt(s) as a comparable soaking medium has been conducted. Given the reduction of cooking times with traditional additives in this study, it may be that their use would reduce soaking time equally, if not faster, than table salt. Further investigation is needed. However, in the interest of eliminating use of traditional salt for health and safety reasons, while still reducing the cooking time for hard-to-cook staple foods like legumes, it is suggested that presoaking be used as an alternative. Adding table salt to the soaking water hastens the process even further, thus reducing the opportunity for bacterial growth. Presoaking provides the benefit of reducing fuel wood use while preventing the consumption of unknown and potentially harmful materials present in traditional additives.

Chapter 5. Palatability of black beans (*Phaseolus vulgaris* L.) cooked in crop ash filtrate and salt additives

5.1. Abstract

Plant ash filtrate is a commonly used additive to cook legumes and other foods in rural Northern Uganda. Ash filtrate is believed to provide a culturally preferred taste as well as reduce cooking time of hard-to-cook dried beans. The aim of this study was to assess the validity of the former, and determine other factors that may also influence preferences. Palatability (sensory) preferences of beans cooked across four treatments (distilled water [control], table salt, ground salt, and ash filtrate) were evaluated through blind taste tests in Northern Uganda. Contrary to anecdotal belief, participants showed an overall preference for black beans cooked with ground salt and table salt over plain beans or those cooked with ash filtrate. Analysis also showed that type of treatment and study site significantly ($p<0.05$) impacted palatability scores, indicating that community culture is influencing taste preference. Demographic and socio-economic factors did not influence palatability preferences within or between communities. Results indicate that cultural preference for the use of ash filtrate is being influenced by more than palatability (actual taste), as in blind testing it was found that ash filtrate was not the preferred treatment for cooking black beans. These findings will provide important support in finding effective methods by which to suggest change among ash filtrate users should the filtrate be found harmful to health or well-being.

5.2. Introduction

Table salt (NaCl) is frequently added to a variety of foods to enhance taste and palatability, and to speed cooking. In Northern Uganda, women may use a combination of commercial table salt and ground salt (precipitate from saline lakes) or plant ash filtrate to infuse a distinct culturally-preferred flavour that is said to be preferred to foods. In particular, these cooking additives are used with hard-to-cook legumes, which benefit from both time saving and improved taste. While subjective and possibly culturally/geographically dependent, sensory acceptability is key to understanding important reasons for either the use or disuse of specific additives in cooking legumes.

Palatability (sensory) evaluation for cooked legumes has previously included colour, taste, texture, and overall acceptability (Silva *et al.* 1981; Onayemi *et al.* 1986; Onwuka & Okala 2003). However, relatively few studies have focused on the use of traditional salts in cooking legumes, and even fewer have included sensory evaluation as a component of the study. Comparison of fresh mucuna beans (*Mucuna pruriens*) cooked with ground salt, citric acid, and plant (maize cob) ash showed preference for the texture of beans cooked with ash and the taste of beans cooked with the ground salt (Wanjekeche *et al.* 2003). Interestingly, taste scores were significantly higher for beans cooked with ground salt (Wanjekeche *et al.* 2003), despite the fact that these two additives share many common properties (Mamiro *et al.* 2011). The colour of both treatments was unacceptable to panelists; a result commonly found with alkaline treatments. Onwuka and Okala (2003) looked at four parameters of palatability for different cooking treatments, including plain water, ground salt (*akanwa*), table salt, and two mixed salts ($NaHCO_3$ and $CaCl_2$). With respect to the first three treatments, respondents rated the legumes cooked with table salt as the most desirable across all parameters, while the legumes cooked with *akanwa* were deemed least acceptable. The colour of beans cooked with *akanwa* was too dark and the flavour less palatable than either the control or beans cooked with table salt and only the texture of the *akanwa* beans was comparable to the other samples. Onayemi *et al.* (1986) also found poor acceptability for cowpeas cooked in either local rock salt (ground salt) or alkali potash (sodium carbonate; functionally similar to plant ash). Prior research using sensory evaluation tests have shown that traditional salts are not preferred, despite a strong cultural presence and continued use. Further, over addition of these salts contributes to a sharp, unpleasant taste (Onayemi *et al.* 1986), possibly due to the high alkalinity. For this reason, bitterness was added as a parameter in this study.

Within these few studies, analysis of palatability preference for common black beans (a staple legume in Uganda) cooked with ash filtrate or ground salt has not been conducted. Also undocumented is the assessment of whether demographic or socio-economic factors may influence preferences. The objective of this study was to determine if beans cooked with ash filtrate are preferred over those cooked in plain water, commercial salt or ground salt. Specifically, palatability preferences were compared for beans cooked in comparative salt treatments. I also examined whether socio-economic factors such as gender, age, or education level influence taste preferences. This will provide information relevant to targeted educational programs should there be any potential adverse health effects. I hypothesized that beans cooked with the ash filtrate would be the most preferred treatment, and that age, gender, and education level would influence overall palatability preferences.

5.3. Materials and methods

5.3.1. Study sites

Beans were cooked near the study sites at Pope Paul Hospital, a central medical center in Atapara Village, Oyam District (36N 431910m E, 247025m N) (Figure 9). Sensory evaluations took place at each of the four study sites; Dog Abam, Telela, and Arok (Oyam District), and Tit (Lira District) in Northern Uganda. A full description of sites can be found in Chapter 2.2.

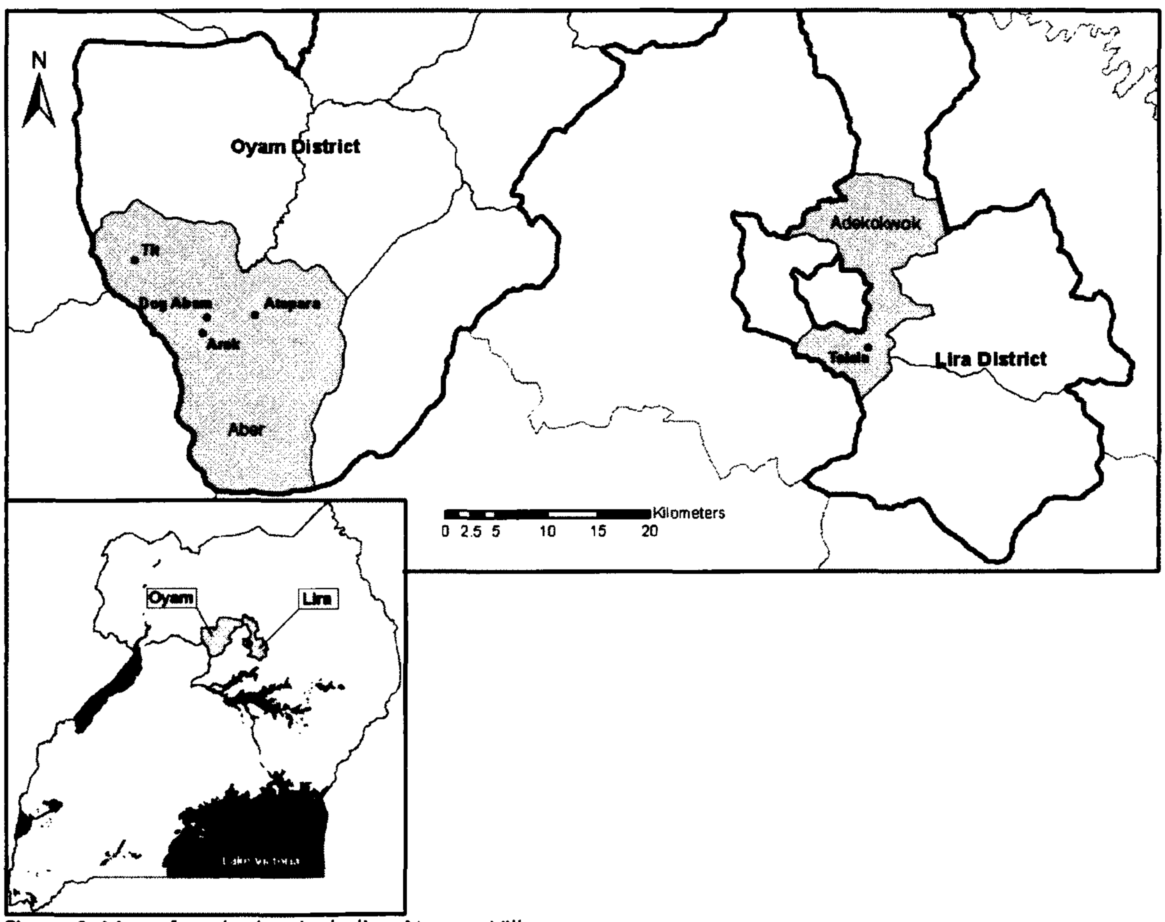

Figure 9. Map of study sites including Atapara Village.

5.3.2. Treatments and sample preparation

The study design consisted of four cooking treatments for black beans (*Phaseolus vulgaris* L.). Four 240 g samples of dried black beans were sorted by hand and shriveled beans and other detritus were removed. Large aluminum pots were thoroughly washed three times prior to use and rinsed with distilled water. Pots were then prepared with the beans, 2.0 litres (L) of distilled water and one of four treatments; distilled water – 'control'; water with 5.7 g of commercial iodized salt – 'table salt', water with 5.7 g of ground salt – 'ground salt', and water with 45 ml (3 tablespoons) of plant ash filtrate – 'ash filtrate'. The plant ash filtrate used for this portion of the study was prepared with ash from the Dog Abam site by a local woman and was documented by photo and video. Ash from Dog Abam was used because there was access to ample supply for use in all sections of this study. The amount of water required for cooking and the treatment concentration was determined by consulting with several local women. These concentrations of table salt (0.23% w/v), ground salt (0.23% w/v), and ash filtrate (1.8% v/v) were also comparable with similar studies (Onayemi *et al.* 1986; Wanjekeche *et al.* 2003). For a full description of additives and method of filtrate preparation, please refer to section 2.4.

Local charcoal stoves were used due to lack of electricity in the region, as well as to replicate typical cooking conditions. Following the traditional cooking methods and learning how to use local equipment, provided an understanding of the time and energy required for the food preparation done daily by local women. Additional distilled water was added as necessary throughout cooking as follows: plain (control) 1.5 L, table salt 1.5 L, ground salt 1 L, and ash filtrate 1.5 L. Beans were tested for completion of cooking by hand

and mouth feel by the author and a local woman employed at Pope Paul Hospital who regularly cooks black beans. This method has been previously used for similar tests (Ankrah & Dovlo 1978; Onwuka & Okala 2003; Wanjekeche *et al.* 2003). Once drained and cooled, each pot of beans was divided into four separate airtight plastic containers for individual use at each site. Each lid was labeled with a code letter and number describing treatment sample and intended site. Samples were refrigerated immediately in a hospital storage refrigerator and all palatability evaluations took place within 36 hours.

5.3.3. Participant selection

The palatability evaluation was completed by 12 people at each study site for a total of 48 participants. Participants were chosen by availability and willingness. Random selection was not an option given logistical and time constraints. There were an equal number of males and females at each site, ranging in age from 18 to 79 years old. Each participant regularly consumed beans prepared with filtrate and so was familiar with the practice of using filtrate for cooking. Signed consent forms were obtained from each person, with an explanation of activities described to them by the research assistant/translator (Appendix D).

5.3.4. Data collection

Each participant answered questions regarding their socio-economic status, including age, gender, education level, and occupation. At each site, a random number draw dictated the order of placement for samples to minimize potential order bias and lids were placed underneath the sample containers to hide the descriptive code from both the participant and research assistant. Evaluations were conducted in private via translation.

Samples were provided by disposable spoon, at ambient temperature. Each treatment was tested using a 5-point Likert type scale for sensory properties of smell, colour, flavour, texture, bitterness (chalkiness), and overall acceptability of beans (Appendix B). Texture was measured on two accounts; both for hardness, from too hard (1) to good (5) and for softness, from too soft (1) to good (5), in order to identify beans which were both deemed to be either under or over-cooked. The scale and categories were adapted from previous successful methodologies (Silva *et al.* 1981; Onayemi *et al.* 1986; Onwuka & Okala 2003).

5.3.5. Informal field interviews

Semi-structured interviews (see Appendix A for list of questions) were conducted to gain local perspective and a cultural context, adding to a more comprehensive understanding of this practice. Due to a lack of census data, interviews were not able to be conducted randomly. Instead, the local research assistant/translator (Ugandan female, aged 40) who was hired to assist with field work acted as a key informant, providing introductions to women representing a variety of demographics. Prior to data collection, a meeting was held with the local research assistant to explain the process and goals of this study, answer any questions, and ensure privacy for all participants through a signed confidentiality agreement (Appendix B). I conducted two trial interviews to clarify and assess appropriateness of questions, determine length of interviews and gain familiarity in working with the research assistant. All interviews (n = 20) were conducted via translation at the homes of each respondent and were recorded on paper.

5.3.6. Statistical analysis

The palatability preference data were collected under the assumption of approximating interval level data (Norman 2010) for analysis with parametric statistical testing. The data did not meet assumptions of normal distribution and primary analysis of treatment means was done through non-parametric measures. Kruskal-Wallis rank tests were performed among treatments with built in post-hoc multiple comparisons to distinguish differing treatments.

Two-factor ANOVA tests were run to further examine whether the site or specific demographic and socio-economic factors (gender, age, and education level) contributed to the treatment effect on overall acceptability of beans. Despite not meeting assumptions of normality, the ANOVA tests were run under the assumption that data would approximate normal as per the central limit theorem (n=48). Statistical significance was determined based on researcher decision at $\alpha = 0.05$. The mathematical model used for the two-factor ANOVA between site and treatment was:

$$Y_{ij} = \mu + \mathit{site}_i + \mathit{treatment}_j + (\mathit{site} \times \mathit{treatment})_{ij} + \varepsilon_{ij}$$

where Y_{ij} = preference (rating for overall acceptability) mean; μ = grand mean preference; site_i = study site (Dog Abam, Telela, Arok, Tit); $\mathit{treatment}_j$ = cooking water treatment (distilled water [control], commercial salt, ground salt, ash filtrate); $(\mathit{site} \times \mathit{treatment})_{ij}$ = interaction between site and treatment; and ε_{ii} = experimental error. The null hypotheses are that there is no difference in means of site, there is no difference in means of treatment, and there is no interaction between site and treatment.

The mathematical models used for the two-factor ANOVAs between demographic and socio-economic factors and treatments were:

$$Y_{ij} = \mu + factor_i + treatment_j + (factor \times treatment)_{ij} + \varepsilon_{ij}$$

where Y_{ij} = preference (rating for overall acceptability)mean; μ = grand mean preference; $factor_i$ = demographic or socio-economic factor of either gender, age, or education level; $treatment_j$ = cooking water treatment (distilled water [control], commercial salt, ground salt, ash filtrate); $(factor \times treatment)_{ij}$ = interaction between demographic or socio-economic factor and cooking water treatment; and ε_{ii} = experimental error. The null hypotheses are that there is no difference in means of any factor (gender, age, or education level), there is no difference in means of treatment, and there is no interaction between the factor (gender, age, or education level) and treatment.

Analysis was completed using the Stata® (Version 12) software package. Compilation of interview responses and determination of percent responses were completed using Microsoft® Excel 2010.

5.4. Results

5.4.1. Palatability preferences among treatments

Statistical analysis showed treatment effects on all sensory characteristics (Table 3). Higher values demonstrate a greater preference for the treatment. There is a distinct preferential pattern for beans cooked with the ground salt treatment across all parameters, followed by beans from the table salt treatment, then beans from the ash filtrate treatment, and least preferred were those cooked in the plain distilled water (control)

(Figure 10). The only anomaly in the rankings was that beans cooked in the ash filtrate scored lowest for bitterness.

Table 3. Chi-squared value, corrected for ties, (x^2, 3 d.f.) and rank mean scores for treatments (n=48). Values along rows with the same superscript are not significantly different at p≥ 0.004167 (adjusted for significance).

		Treatment			
Characteristic	x^2	**Control**	**Table Salt**	**Ground Salt**	**Ash Filtrate**
Smell	18.13 (p<0.001)	77.13^{a}	107.00^{ab}	117.98^{b}	83.90^{a}
Colour	12.04 (p=0.007)	79.09^{a}	106.03^{ab}	111.18^{b}	89.70^{ab}
Flavour	27.55 (p<0.001)	70.61^{a}	116.30^{b}	116.83^{b}	82.25^{a}
Texture (hardness)	33.60 (p<0.001)	58.91^{a}	107.35^{b}	119.28^{b}	100.46^{b}
Texture (softness)	29.38 (p<0.001)	61.65^{a}	106.68^{b}	116.45^{b}	101.23^{b}
Bitterness	12.10 (p=0.007)	88.09^{ab}	102.52^{ab}	115.18^{a}	80.21^{b}
Overall	31.41 (p<0.001)	68.80^{a}	112.44^{b}	122.74^{b}	82.02^{a}

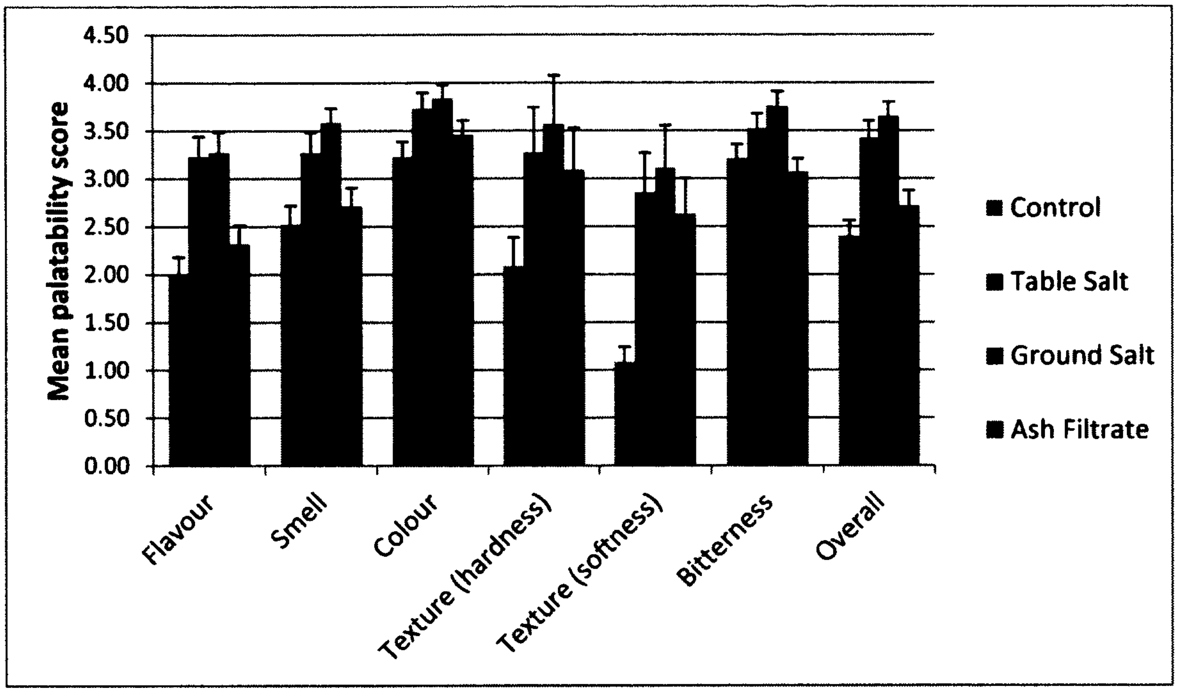

Figure 10. Mean (± standard error) scores of palatability preference by parameter for treatments.

Several two-factor ANOVAs were used to identify possible interactions between study site, treatment and three socio-economic factors; age, gender, and education level. Outcomes revealed a significant ($p<0.05$) main effect of both treatment and study site on overall palatability preference, suggesting cultural taste variation on a small geographic scale. There was also a significant interaction between the two factors, with specific differences evident at the Arok site, where the table salt treatment was not well accepted (Figure 11). Otherwise, the general trend of taste preferences supported Kruskal-Wallis results for overall preference scores. Further independent variables investigated with cooking treatment through two-factor ANOVA included factors of gender, age, and education level. There was no main effect of gender, age, or education level, however the treatment type was a significant ($p<0.05$) main effect in each test. There were no interactions between treatment and any of gender, age, or education level.

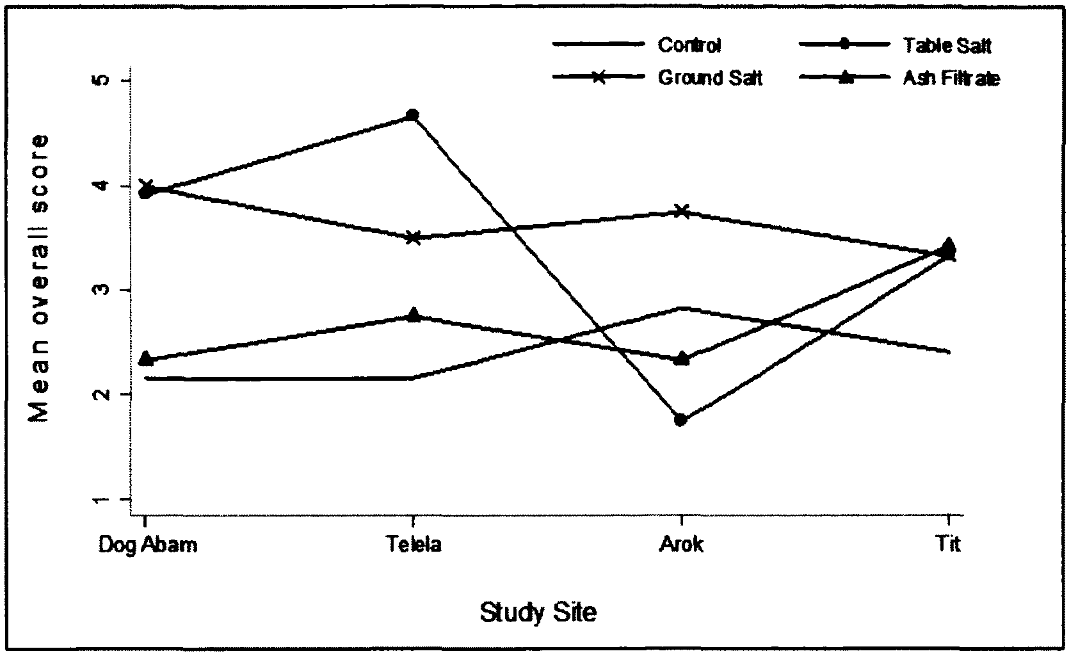

Figure 11. Interaction terms between study site and treatment type on respondents' overall palatability scores.

5.4.2. Participant responses to ash filtrate use and cultural importance

During the interviews, 85% of women stated that they preferred beans cooked with ash filtrate for improved taste, smell, or both, which did not correspond to the statistical findings of the blind taste tests. Two women answered that they did not prefer to use it but that it was necessary, "For me, it is the culture that makes me use it but I don't like it". There was one nonresponse. Several respondents said that they add table salt and other ingredients (e.g., peanuts) to the beans after they are cooked with filtrate, to further improve flavour.

5.5. Discussion

Sensory evaluation across study sites yielded surprising results given anecdotal beliefs and interview responses. Beans cooked in ash filtrate were not well scored and almost exclusively the second to last preferred of all treatments. The sole parameter which rated beans cooked with ash filtrate over that of the control was improved texture (Table 3), probably due to the effect of alkalinity on breakdown of legume cell properties (Ankrah & Dovlo 1978; Varriano-Marston & de Omana 1979; Uzogara *et al.* 1990; Onwuka & Okala 2003). However, the high alkalinity of the ash filtrate treatment was likely the reason why beans cooked with the ash filtrate scored lowest for bitterness. Also unexpected was the preference for the ground salt treatment as prior research results indicated that it contributed to an unpleasant colour (Wanjekeche *et al.* 2003) and flavour (Onayemi *et al.* 1986; Onwuka & Okala 2003). Addition of ground salt can cause beans to turn a darker, less appealing colour which has resulted in low acceptability (Onayemi *et al.* 1986; Wanjekeche *et al.* 2003). The same results were not found in this study, perhaps because black beans were used and the colour variability was less apparent than with the lighter coloured beans used previously. The flavour however, was clearly preferred in the ground salt treatment and the table salt treatment. Both of these treatments contain higher sodium content than the other treatments (Table 9, Chapter 6), suggesting that this is an important contributor to sensory acceptability.

In comparison of study sites, the ash filtrate treatment was not the most preferred treatment at three of the four sites, and scored highest (although with no statistical significance and by a very small margin over the ground and table salt treatments) by respondents only at Tit village (Figure 11). All other sites rated beans cooked with ash filtrate very poorly, and only slightly better than those cooked in distilled water. Participants at the Dog Abam site preferred beans cooked in both salt treatments by a large margin over those cooked in the control and ash filtrate treatments, suggesting a greater affinity for sodium. Respondents from Telela showed a strong preference for beans cooked in table salt. This village is located closest to a large town (Lira) and it may be that availability of table salt is easier, thus developing a preference for its use, or that traditional customs are somewhat less relied upon, thus lessening the preference for ground salt and ash filtrate. In contrast, respondents from the Arok study site greatly disliked the table salt treatment compared to participants from all other sites (Figure 11). The reason for this was not apparent, other than local preferences, as this study site was located not far from Dog Abam and no observable differences in these study sites were identified during data collection. However, the difference may be due to factors not identified in this study. Participants from Tit showed preference for any cooking additive over the control, which may be a function of the villages' history. This village was the last of the four study sites to resettle after living at an Internally Displaced Persons (IDP) camp for many years. Scarcity of either table salt or traditional additives during those years may have subsequently made the availability of any additive welcome.

The importance of analyzing sensory preferences lies in its relevance to the culturally dominant and accepted belief that cooking beans with ash filtrate provides the best taste. If there should be any health or environmental concerns identified, the fact that people preferred beans not cooked with ash filtrate would be important in developing an educational plan about this practice. The results of statistical tests showed that both site and treatment were important in understanding differences in preferences, as was the interaction between them. It was anticipated that treatment would affect sensory evaluation, but it is interesting that site (location) also affected preference. This suggests important cultural connections within small populations, which makes sense as women within these communities often share cooking duties and materials, such as ash. These women and their families may develop similar preferences for food. An important lesson from this finding is that there are different and complex traditions (e.g., cooking methods and/or amount of ash filtrate added) to consider among even small communities, that influence individuals' preferential tastes for food. It would be inaccurate to treat these communities as homogenous by assuming similar taste preferences throughout.

None of the socio-economic factors were found to be of statistical significance in contributing to observed differences among treatments. However, when interpreting these findings in a real life context, it must be noted that gender is important, as women perform a vast majority of domestic duties and as such, are responsible for how meals are prepared (Madge 1994). The perceived benefits of using ash filtrate include improved taste and smell of food, as well as decreased cooking time for hard-to-cook foods. The distinct flavour imparted by the filtrate has resulted in it being used ubiquitously by households, and subsequently becoming a culturally obligatory practice. The practical benefit of decreased cooking time reduces daily workload for women, by reducing the number of hours of meal preparation and the time spent on associated activities (e.g., collection of fuel wood). In the event that the consumption of ash filtrate is found to be nutritionally harmful, it would be the women with whom potential alternative methods of cooking would be discussed. The results of the blind taste tests in this study may be used to identify preferences between study sites and form a basis from which to initiate an individualized conversation with women from each community.

Several factors which may have influenced the dislike for beans cooked with ash filtrate in this study need to be considered. While every attempt was made to replicate common cooking practices, women in this region are used to preparing their own food and so have personal preferences that cannot be duplicated in every case (e.g., such as amount of filtrate to add and how long to cook the beans). Those interviewed indicated that the over-addition of filtrate creates very bitter tasting beans, and as the filtrate treatment scored poorly for bitterness (Table 3) it may be that the amount used was not in accordance with all household preferences. However, low scores across most parameters for the beans cooked in ash filtrate treatment suggest a general dislike. Also, refrigeration is very rarely used in this area and while refrigerated samples were warmed to ambient temperature before testing, this may have impacted taste. Lastly, cooked beans are always served with a sauce made from the remains of cooking water and addition of peanuts, salt, and/or other additives. The retention of this sediment likely provides a large portion of the flavour and so the taste differences between drained bean samples would be very subtle. In a region with very limited exposure to novel foods or cooking practices, participants may have had difficulty in differentiating specific sensory parameters in an atypically cooked food such as plain dry beans.

Legumes cooked with ash filtrate, while identified as being culturally preferred, did not rank well with legumes cooked with ground salt and table salt in blind testing. This information can be used to strengthen the case against the use of ash filtrate, should it be shown to be potentially harmful. Given the contradictory findings among study sites, as well as between this research and other studies, sensory evaluations pertaining to specific populations or areas should always be conducted to identify area-relevant preferences.

Chapter 6. Composition of ash residue and potential health impacts of ash filtrate used for cooking dried black beans (*Phaseolus vulgaris* L.)

6.1. Abstract

Ash from burnt crop residue of *Phaseolus vulgaris* (L.) is commonly used to generate filtrate in rural Northern Uganda. The filtrate is added to hard-to-cook foods, like dried legumes, to decrease cooking time and improve flavour. However, the composition of ash filtrate and the health implications of its use are poorly understood. The purpose of this study was to determine the elemental contents of the crop ash and ash filtrate, identify variation among study sites, and assess the impact of ash filtrate on nutritional health. Dried ash samples of *P. vulgaris* collected from Dog Abam, Telela, Arok, and Tit villages in Northern Uganda were subjected to inductively coupled plasma – mass spectrometry (ICP-MS) analysis to estimate the chemical composition. Results from crop residue ash showed significant differences in potassium, sodium, magnesium, iron, zinc, manganese, arsenic, silicon, mercury, and titanium levels among sites. Elemental contents of ash filtrate are much lower than crop residue ash. The commonly-used table salt in the study areas had significantly higher sodium but lesser potassium contents than crop residue ash. Elemental concentration present in probable daily intake of ash filtrate through cooked beans (approximately 15 milliliters/person) were within the acceptable range for recommended daily intake according to the Ugandan and Canadian nutritional standards. However, the alkaline pH levels of the ash filtrate (pH 10.1 to 10.8) may contribute to anti-nutritional effects by decreasing the bioavailability of specific minerals (e.g. iron and zinc) and/or having deleterious effects on various nutrients (e.g. thiamine). Given the potential negative effect

of ash filtrate on diet, alternative methods to decrease cooking time for dried legumes, such as pre-soaking, are proposed in the study areas. Detailed nutritional evaluation of the health of individuals should also be conducted to identify specific negative impacts of the frequent consumption of ash filtrate.

6.2. Introduction

In Northern Uganda, ash is obtained from burnt crop residue and dried plant parts to produce a concentrated filtrate. Knowledge of local women states that filtrate, when added to the cooking water of hard-to-cook foods (e.g., dried legumes), accelerates cooking time and adds a culturally preferred flavor to the dish. However, the effect of ash filtrate on health through its consumption is unknown. Based on information about the elemental concentration in dry plant ash from previous studies (Calloway *et al.* 1974; Kuhnlein *et al.* 1979; Kuhnlein 1980), there is concern that the ash filtrate may also contain high levels of elements that may be harmful to consumers' health. Burning crop residues will vaporize volatile or biological compounds (Adriano 2001), and concentrates any plant elements in the form of ash (Townsend *et al.* 1973). Further, the legumes (at least in Northern Uganda) are usually burnt with the attached soil which may also contribute elements to the ash mixture. Ferralsols, the dominant soils in this region, are acidic with high contents of iron and aluminum (Eswaran *et al.* 1997; ISRIC 2013; Opio 2013), and as such their enrichment may be of particular concern in the crop residue ash.

The crops most commonly used for making ash filtrate in Northern Uganda are legumes, which includes the soy, black, kidney, and yellow varieties of the common bean (*Phaseolus vulgaris* L.). Sesame (*Sesamum indicum*) and peanuts (*Arachis hypogaea*) are also used, though less frequently. *P. vulgaris* varieties are often mixed during drying and burning, with little preference for one variety over another (A. Akello, pers. comm. 2012). This suggests that comparable cooking and taste effects are attained from all above listed varieties.

Plant parts have been used as a traditional type of salt additive in other parts of Africa (Culwick 1950; Huntingford 1955; Kaputo 1996; Wanjekeche *et al.* 2003; Mamiro *et al.* 2011), Papua New Guinea (Townsend *et al.* 1973; Ohtsuka *et al.* 1987), and the United States (Calloway *et al.* 1974; Kuhnlein *et al.* 1979; Kuhnlein 1980). The use of *P. vulgaris* in several regions includes dry ash, either eaten alone as a nutritional supplement or added to other food preparation. The Hopi American Indian tribe used a 'culinary ash' usually made by burning salt bush (*Atriplex canescens*) and/or bean (*P. vulgaris*) debris (Calloway *et al.* 1974; Kuhnlein *et al.* 1979; Kuhnlein 1980). In dry ash, the levels of iron, lead, and strontium were of particular concern, especially in the bean ash sample (Calloway *et al.* 1979) where lead (11 ± 2 mg/kg) and strontium (1870 ± 190 mg/kg) exceeded the regulated tolerance levels in foods. More recent analysis of essential elements present in *P. vulgaris* ash in Africa has shown varying levels of several minerals. For example, in Zambia, iron was reported at 1050 ± 0.51 parts per million (ppm), copper at 24 ± 1.13 ppm, and potassium present at 65 ± 8.25 % (Kaputo 1996). In Tanzania, *P. vulgaris* ash contained 41 ± 3 ppm of iron, 4 ppm of copper, and 650 ± 109 ppm of potassium (Mamiro *et al.* 2011). To date, there has been no available data on the total elemental composition of common plant ash used in Uganda for traditional food preparation. There has also been no elemental analysis of plant ash filtrate, or investigation into the contributions of dry ash or filtrate to the daily mineral intake levels in the diet. Further, it has been presumed that bean crop ash contains high levels of sodium given its use as a traditional salt replacement; however previous studies have been inconclusive (Kaputo 1996; Mamiro *et al.* 2011). This study provides a comprehensive elemental compositions of crop ash filtrate, dry crop ash and salts (ground salt and commercial salt) used in traditional food preparation in Northern Uganda. The potential dietary nutritional impact of the daily use of ash filtrate was also explored.

The objective of this study was to investigate any potential general health impacts of ash filtrate and make recommendations about its use in food preparation through: 1) comparison of the elemental contents of the crop ash and crop ash filtrate between study sites, transfer of elements from crop ash to filtrate and comparison of crop ash against salts commonly used in the region; 2) comparison of the contributions from beans cooked with filtrate, ground salt, and table salt against the daily recommended mineral intake; and 3) discussion of potential health implications based on current practices of ash filtrate use in Northern Uganda. It was my hypothesis that elemental concentration in plant ash or ash filtrate would not be significantly different among sites, given that the same crop species are used for the same functional purposes across a relatively widespread area. It was also my hypothesis that there would be significant differences in sodium and potassium levels between salts and plant ash. Lastly, I presumed that ash filtrate could be negatively affecting the diet of users through the contribution of high levels of required and/or toxic elements.

6.3. Materials and methods

6.3.1. Sample collection and filtrate extraction

Four 150 g ash samples of the common varieties of *P. vulgaris* were collected from each of Dog Abam, Telela, Arok, and Tit study areas in Northern Uganda (see Figure 2, Chapter 1). Two ash samples were collected from each of two households at each study site (n=4 x 2 x 2=16) generated from the 2011 and 2012 harvest years. While the intent was to collect individual random samples, lack of census data about households and access issues prevented this. However, it was observed that harvest and ash burning are community activities. The ash is shared among several families within villages, precluding the necessity to analyze variation among a village or community. The dried ash was collected using a plastic scoop and stored in a 250 millilitre (ml) wide mouth high-density polyethylene (HDPE) container. Two 500 g bags of ground salt (precipitate from saline lakes) from the Lake Magadi area in Kenya were purchased from vendors at the Lira Market. A 200 g sub-sample of salt was drawn from each bag and stored in 250 ml HDPE container for total elemental analysis. A 500 g bag of Chiluma iodized commercial sea salt (Kensalt Limited in Kenya) was purchased in Kampala. One 200 g sample was drawn, sealed in a 250 ml wide mouth HDPE container. These samples were stored dry and without preservative. Each container was sealed and labeled with date, study site, sample identification number, year of procurement, and geographical coordinates for transport to Canada. A full description of ash procurement is provided in Chapter 2.4. Sub-samples of dry ash and salts (approximately 5 grams each) were separated in the laboratory at the University of Northern British Columbia (UNBC) into sterile 20 ml glass scintillation vials (n=16) for the determination of elemental contents using inductively coupled plasma - mass spectrometry (ICP-MS) analysis (see below).

The ash filtrates were prepared at UNBC using a method as similar as possible to the method employed by the local women in the study areas in Northern Uganda. Ash samples from the same household at each study site were combined to meet the amount necessary to generate ash filtrate (n=4 from each study site became n=2 from each study site except from Telela where n=4 became n=1 due to lack of supply) for a total of 7 filtrate samples. Approximately 250 g of loose, dry ash was placed into an unbleached coffee filter to mimic the rudimentary spear grass filter typically used in the study areas. The filter with ash was placed into a perforated (0.5 cm holes at the bottom) plastic cup from Uganda. The cup was washed and rinsed with Type 1 water[2], and dried prior to use and in-between samples to prevent any cross contamination. A small portion of 200 ml of Type 1 water was added to the ash to form a filter bed. The remainder of the water was slowly added and percolated through the ash and collected in another plastic cup as ash filtrate. Filtrate samples (n=7) were stored without preservatives in sterile 20ml glass scintillation vials prior to the determination of elemental contents using ICP-MS (see below). The pH of each filtrate sample was taken using a Bluelab® pH pen. Values of pH were recorded for each treatment once the reading held steady for 10 seconds, and pH pen was rinsed in Type 1 water prior to subsequent test.

Sample collection of beans cooked in each treatment was done following the time trials described in Chapter 4 (n=4). Wet samples (approximately 10g) of drained cooked beans were collected in sterile 20 ml glass scintillation vials from each pot after beans were cooked to a 150 g puncture force. Beans were not treated with any preservative and were submitted immediately to laboratory for ICP-MS analysis (see below).

6.3.2. Elemental analysis

All laboratory analysis was completed at the Central Equipment Laboratory at UNBC by a certified technician. Crop ash, salt and bean samples (approximately 0.4 g each) were

[2] Ultrapure water filtered with a Millipore® system meeting ASTM® standards for Type 1 water.

oven-dried at 70-80° C overnight (48 hours for bean samples), and then microwave digested with 4 ml concentrated nitric acid following the methods suggested for organic ash samples by the United States Environmental Protection Agency (EPA Method 3052) (US EPA 2014a). Microwave digested ash and bean samples were heated to 230°C for ten minutes and diluted to 50 ml with Type 1 water. Moisture content of air-dried beans was determined by oven-drying at 60°C for 24 hours. Aqueous filtrate samples (9 ml) were acidified with 1 ml of nitric acid, heated on a 10 minute ramp to 180°C, held at 180°C, and then diluted to 50 ml with Type 1 water (following EPA3015A method) (US EPA 2014b). Samples were analyzed with ICP-MS (Agilent 7500Cx) for the following elements: lithium, beryllium, boron, sodium, magnesium, aluminum, silicon, phosphorus, potassium, calcium, titanium, vanadium, chromium, manganese, iron, cobalt, nickel, copper, zinc, gallium, germanium, arsenic, selenium, rubidium, strontium, zirconium, niobium, molybdenum, silver, cadmium, tin, antimony, tellurium, cesium, barium, lanthanum, cerium, praseodymium, neodymium, samarium, europium, gadolinium, dysprosium, holmium, erbium, thulium, ytterbium, lutetium, hafnium, tantalum, tungsten, mercury, thallium, lead, bismuth, thorium, and uranium. From the above list, subsets of elements were further investigated based on the nutritional guidelines for Canada and Uganda (National Drug Authority 2009; Health Canada 2010). Macro elements are dietary minerals needed in amounts over 100 mg/day while micro elements are required in much lower quantities (Kirch 2008). Additional elements were included in the analysis because they are potentially required, of unknown necessity, or have been suggested as potentially harmful elements (WHO 1996; Trumbo *et al.* 2001).

6.3.3. Sample calculations

The daily elemental intake from filtrate was calculated as follows:

*Daily elemental intake from filtrate (mg) = elemental concentration in filtrate (ng/ml) * 15 ml * 1 mg / 1000000 ng* (1)

where a daily intake of 15ml represents the typical amount of filtrate used to cook the commonly eaten dried legumes in the study areas.

Daily intake of elements from bean consumption was calculated using the following relationship:

*Daily intake of element from dried beans (mg) = elemental concentration in oven-dried bean values (mg/kg) * 0.077 kg oven-dried bean* (2)

where 0.077 kg is equivalent to an 80 g portion of air-dried beans typically consumed on a daily basis in the study areas. A moisture content of 3.9% was used in the calculations based on results of moisture content analysis of beans used in the study.

To determine the proportion of an element extracted from the ash, the following equations were used:

*Proportion (%) = (total elemental amount in 200 ml filtrate (mg) / total elemental amount in 250 g ash (mg))*100* (3)

where total elemental amount in 250 g ash is the quantity of ash used to produce filtrate (i.e., elemental concentration in oven-dried weight (mg/kg) * 0.25 kg = mg of element) and total elemental amount in 200 ml filtrate is the volume of Type 1 water used to produce filtrate (i.e. elemental concentration in filtrate (mg/l) * 0.2 l = mg of element). Average elemental values for dry ash (n = 16) and filtrate (n = 7) for each study site were used for the calculations. The proportion of extracted elements shows the percentage of transferred elements from oven-dried ash to filtrate through the filtrate procurement process.

6.3.4. Informal field interviews

Semi-structured interviews (see Appendix A for list of questions) were conducted to gain cultural context and knowledge towards a comprehensive understanding of the practice of using ash filtrate in food preparation. Due to a lack of census data, interviews were not randomly conducted. Instead, a local research assistant/translator (Ugandan female, aged 40) who was hired to assist with field work acted as a key informant, providing introductions to women representing a variety of demographics. Prior to data collection, a meeting was held with the local research assistant to explain the process and goals of this study, answer any questions, and ensure privacy for all participants through a signed confidentiality agreement (Appendix B). I conducted two trial interviews to clarify and assess appropriateness of questions, determine length of interviews and gain familiarity in working with the research assistant. All interviews (n = 20) were conducted via translation at the homes of each respondent and were recorded on paper.

6.3.5. Statistical analyses

Two assumptions were made prior to the statistical analysis of the elemental contents of the samples. First, that harvest year of ash generation was not a principal factor because ash is used up to and past the harvest year. Second, that ash does not differ between households because neighbours often share ash burning duties and therefore, the produced ash. Households included in this study were not randomly chosen for this reason and due to lack of census data and time constraints.

Data was tested for normality using the skewness and kurtosis test. When assumptions were not met, data transformations were made. Homogeneity of variance was confirmed by Levene's statistic. Where a statistically significant ($\alpha = 0.05$) effect of site was found, post-hoc Tukey's Honestly Significant Difference (HSD) multiple comparison tests were used to determine significant differences among treatment means (Gotelli & Ellison 2004). The null hypothesis was that there would be no difference among elemental means from each study site.

Analysis of variance (ANOVA) was used to compare elemental concentration in ash samples among sites. The mathematical model used was:

$$Y_i = \mu + site_i + \varepsilon_i$$

where Y_i = elemental concentrations (see Table 4 for elements included in statistical analysis); μ = grand mean element, $site_i$ = study site (Dog Abam, Telela, Arok, Tit); and ε_i = experimental error.

T-tests were used to compare sodium and potassium contents between combined salts (ground and commercial salt) and plant ash samples. Skewness and kurtosis tests for normality were not met for the samples but data was successfully transformed. Homogeneity of variance was confirmed by Levene's test. Statistical significance was determined at $\alpha = 0.05$ and the null hypothesis was that no difference of means existed between salt and ash.

Statistical analysis was done using Stata® (Version 12). Compilation of interview responses and summary statistics of pH values were completed using Microsoft® Excel 2010.

6.4. Results

6.4.1. Elemental contents in ash, ash filtrate, and salts

Elemental contents of oven-dried crop residue ash samples varied statistically significantly ($p<0.05$) among sites (Table 4), for some elements. Range of elemental contents (mg/kg) included potassium (88188 - 165577), sodium (148 - 435), magnesium (19803 - 35529), iron (3644 - 10729), zinc (83 - 179), manganese (326 - 963), arsenic (0.38 - 0.70), silicon (244 - 437), and titanium (111 - 290). Elements in ash samples collected at the Arok site were generally significantly lower than other sites. Ash samples from the Dog Abam site showed generally higher concentrations of elements than other sites, although not often statistically significant. Manganese contents varied the most statistically among sites ($p<0.05$), but the greatest range in values was for potassium levels (165,577 mg/kg at Dog Abam to 88,188 mg/kg at Arok). However, given the relatively modest variation among sites in general, and particular the similarity of the ash samples from Telela to the other sites despite the distance separating it (Figure 9, Chapter 5), it can be deduced that elemental concentration is fairly similar among ash of *P. vulgaris* in the geographical region of this study.

Comparison between salts typically used in Northern Uganda and plant ash samples showed that sodium levels were significantly higher in salts (251261 ± 58558 mg/kg) than in plant ash (243 ± 33 mg/kg) (Table 5). Conversely, potassium levels were significantly higher in plant ash (139236 ± 8827 mg/kg) than in salts (11121 ± 5445 mg/kg).

Table 6 displays the amounts of elements (as a percentage) extracted from crop residue ash and transferred into the ash filtrate. The proportion of extracted molybdenum (6 - 35%), silicon (9 - 19%), sodium (6 - 9%), potassium (5 - 7%), and rubidium (5 - 7%) were considerably greater than all other elements. Overall however, it appears that much of the elemental concentration in ash is retained and not leached into the ash filtrate.

The filtrate sample from Dog Abam showed higher concentration for most elements (e.g. calcium, phosphorus, iron, and manganese, in particular) and was lower only in magnesium (Table 7). It was also highly alkaline at 10.8 ± 0.1 (Table 7). There is also much higher variation within this site, reflective of a greater difference between filtrate samples from this site. A typical daily consumption amount of approximately 15 ml of filtrate shows that very little amount of elements is being consumed through the addition of ash filtrate (Table 8).

Elemental contents of beans cooked in each treatment are shown in Table 9. The information from this table was used in calculating daily serving amounts of beans for Table 10. As calculated, Table 10 displays the elemental concentration in daily serving (80 g) of beans cooked in each treatment, set against recommended dietary intake (RDI) tables for adults. Differences among treatments are generally not pronounced, with the exception of higher sodium levels in the table salt and ground salt treatments. The ground salt treatment also had higher amounts of iron, aluminum, and titanium. Boron, aluminum, and strontium were the only elements which exceeded set upper limits of RDI. However, contents of several other elements including nickel, rubidium, barium, and titanium could not be assessed based on lack of sufficient data for regulations.

Table 4. Mean (± standard error) contents elements in ash samples from Dog Abam, Telela, Arok and Tit sites in Northern Uganda (n=16). Values along rows with the same superscript are not significantly different (p > 0.05) by the Tukey's HSD method.

Element		**Crop Ash Residue**			
		Dog Abam *(mg/kg)*	**Telela** *(mg/kg)*	**Arok** *(mg/kg)*	**Tit** *(mg/kg)*
Calcium	Ca	77626 ± 2290^{a}	101276 ± 1258^{a}	77062 ± 13734^{a}	86943 ± 1753^{a}
Phosphorus	P	13503 ± 1197^{a}	12157 ± 166^{a}	8944 ± 1718^{a}	8071 ± 635^{a}
Potassium	K	165577 ± 5272^{a}	143377 ± 1253^{ab}	88188 ± 17070^{b}	159803 ± 932^{ab}
Sodium	Na	435 ± 73^{a}	221 ± 3^{a}	148 ± 5^{b}	168 ± 4^{ab}
Magnesium	Mg	35529 ± 1759^{a}	21525 ± 185^{b}	19803 ± 2645^{b}	29782 ± 873^{ab}
Iron	Fe	3644.89 ± 26.28^{a}	6188.04 ± 165.98^{a}	7472.46 ± 949.15^{ab}	10729.05 ± 482.74^{b}
Zinc	Zn	179.85 ± 19.50^{a}	142.83 ± 1.62^{ab}	88.48 ± 11.43^{ab}	83.62 ± 2.35^{b}
Manganese	Mn	963.10 ± 113.29^{a}	326.26 ± 5.28^{b}	458.11 ± 6.03^{c}	543.62 ± 12.67^{ac}
Copper	Cu	51.95 ± 4.03^{a}	95.04 ± 1.79^{a}	78.92 ± 16.94^{a}	48.19 ± 2.24^{a}
Selenium	Se	1.12 ± 0.04^{a}	0.84 ± 0.03^{a}	0.97 ± 0.10^{a}	1.21 ± 0.03^{a}
Molybdenum	Mo	0.44 ± 0.03^{a}	2.00 ± 0.06^{a}	1.17 ± 0.28^{a}	0.49 ± 0.03^{a}
Chromium	Cr	14.42 ± 0.18^{a}	8.90 ± 0.20^{a}	23.64 ± 3.68^{a}	16.17 ± 2.27^{a}
Nickel	Ni	36.79 ± 7.78^{a}	7.64 ± 0.14^{a}	11.04 ± 1.35^{a}	9.00 ± 0.37^{a}
Cobalt	Co	19.38 ± 4.25^{a}	3.23 ± 0.09^{a}	4.18 ± 0.70^{a}	4.03 ± 0.12^{a}
Arsenic	As	0.38 ± 0.01^{a}	0.40 ± 0.01^{ab}	0.60 ± 0.07^{ab}	0.70 ± 0.03^{b}
Boron	B	273.58 ± 2.15^{a}	245.64 ± 2.40^{a}	169.89 ± 26.09^{a}	277.75 ± 9.81^{a}
Silicon	Si	437.01 ± 13.22^{a}	255.86 ± 5.15^{b}	244.01 ± 9.32^{b}	244.61 ± 6.08^{b}
Vanadium	V	13.91 ± 0.36^{a}	13.29 ± 0.34^{a}	20.21 ± 2.66^{a}	23.81 ± 0.95^{a}
Rubidium	Rb	137.78 ± 17.29^{a}	91.36 ± 0.76^{a}	121.85 ± 28.42^{a}	94.53 ± 5.72^{a}
Strontium	Sr	641.36 ± 4.80^{a}	1064.40 ± 12.87^{a}	747.15 ± 148.39^{a}	619.73 ± 15.71^{a}
Barium	Ba	1591.71 ± 140.05^{a}	1068.51 ± 11.60^{a}	804.99 ± 104.43^{a}	770.25 ± 63.63^{a}
Aluminum	Al	11194.47 ± 81.15^{a}	13705.95 ± 375.06^{a}	13606.70 ± 1843.47^{a}	16596.04 ± 756.62^{a}
Lead	Pb	3.28 ± 0.15^{a}	2.39 ± 0.05^{a}	4.99 ± 0.96^{a}	3.78 ± 0.39^{a}
Mercury	Hg	0.01 ± 0.0004^{a}	0.01 ± 0.0006^{a}	0.01 ± 0.0003^{a}	0.02 ± 0.002^{a}
Titanium	Ti	205.71 ± 18.09^{ab}	290.93 ± 8.12^{a}	111.50 ± 8.10^{b}	285.61 ± 9.26^{a}

Table 5. Mean (± standard error) contents of sodium and potassium in combined salts (n=3) and plant ash samples (n=16). Values along rows with the same superscript are not significantly different (p>0.05).

Element	Plant Ash *(mg/kg)*	Combined Salts *(mg/kg)*
Sodium	243 ± 33[a]	251261 ± 58558[b]
Potassium	139236 ± 8827[a]	11121 ± 5445[b]

Table 6. Mean proportion (percent) (± standard error) of elements extracted from plant ash samples in filtrate procurement. Values were calculated from averages by study site for plant ash (n=16) and ash filtrate (n=7), from Dog Abam, Telela, Arok and Tit sites in Northern Uganda. A single measure of ash filtrate was available for Telela.

Element	Study Site						
	Dog Abam		Telela	Arok		Tit	
Ca	0.12	±0.04	0.05	0.07	±0.03	0.06	±0.01
P	0.18	±0.07	0.02	0.05	±0.03	0.03	±0.01
K	6.41	±1.52	4.93	6.65	±4.63	7.16	±0.22
Na	8.71	±4.17	6.08	6.30	±0.66	7.97	±1.67
Mg	0.03	±0.02	0.64	0.67	±0.31	0.34	±0.34
Fe	0.01	±0.01	0.00	0.00	±<0.0001	0.00	±<0.0001
Zn	0.02	±0.02	0.00	0.00	±0.001	0.00	±<0.0001
Mn	0.01	±<0.01	0.00	0.00	±0.003	0.00	±<0.0001
Cu	0.06	±0.04	0.02	0.02	±0.02	0.02	±0.01
Se	0.42	±0.21	0.60	0.61	±0.39	0.44	±0.06
Mo	16.77	±9.76	7.19	6.08	±5.49	34.58	±21.47
Cr	1.16	±0.75	0.24	0.02	±0.01	0.19	±0.19
Ni	0.03	±0.02	0.01	0.12	±0.12	0.05	±0.04
Co	0.03	±0.02	0.02	0.11	±0.08	0.03	±0.008
As	1.06	±0.71	0.07	0.09	±0.05	0.21	±0.13
B	4.95	±1.13	1.32	1.21	±0.56	2.45	±0.38
Si	18.60	±6.87	9.30	8.70	±1.33	12.59	±3.00
V	1.45	±0.52	0.31	0.07	±0.03	0.15	±0.09
Rb	6.24	±2.76	4.99	6.60	±6.68	4.75	±1.51
Sr	0.03	±0.02	0.02	0.03	±0.02	0.01	±0.007
Ba	0.02	±0.01	0.01	0.02	±0.01	0.01	±0.004
Al	0.02	±0.01	0.00	0.00	±<0.0001	0.00	±<0.0001
Pb	0.07	±0.06	0.01	0.00	±0.001	0.00	±<0.0001
Hg	0.91	±0.53	0.33	0.46	±0.08	0.14	±0.08
Ti	0.02	±0.02	0.00	0.00	±<0.0001	0.00	±<0.0001

Table 7. Mean (± standard error) values of pH and elemental concentrations of ash filtrate samples from Dog Abam, Telela, Arok and Tit sites in Northern Uganda (n=7; n=2 for each study site except for Telela where n=1). The blank sample is an average of duplicate Type 1 water samples.

	Blank		Crop Ash Filtrate						
			Dog Abam		Telela	Arok		Tit	
pH			10.8	±0.1	10.1	10.5	±0.1	10.3	±0.4
Element	(ng/ml)		(ng/ml)		(ng/ml)	(ng/ml)		(ng/ml)	
Ca	121	±81.64	118353	±34161	60728	65119	±2733	65988	±4862
P	<5	±<0.001	29833	±9717	2652	5061	±672	3278	±598
K	26	±2.01	13275574	±2910205	8844479	7330373	±3159224	14302324	±360327
Na	1025	±4.85	47390	±2747	16820	11665	±250	16742	±3361
Mg	1	±0.23	12188	±11901	172794	166706	±41820	125174	±124524
Fe	0.77	±0.07	407.77	±397.91	5.02	12.49	±0.20	10.18	±7.14
Zn	0.41	±0.06	55.10	±52.11	1.95	2.47	±0.26	1.13	±0.18
Mn	<0.01	±<0.001	142.23	±140.41	0.95	19.19	±18.38	26.52	±25.96
Cu	0.11	±0.004	37.98	±24.24	17.87	20.50	±14.91	13.07	±8.78
Se	0.32	±<0.001	5.90	±2.82	6.29	7.40	±4.19	6.60	±0.82
Mo	0.31	±0.08	91.66	±49.93	179.87	89.13	±54.32	210.51	±123.90
Cr	0.41	±0.024	209.94	±135.42	26.78	5.00	±3.87	37.64	±36.20
Ni	0.02	±0.014	15.63	±2.71	0.51	17.15	±14.92	5.26	±4.68
Co	<0.01	±<0.001	8.11	±3.65	0.84	5.79	±3.60	1.28	±0.39
As	0.04	±0.032	5.06	±3.38	0.35	0.65	±0.33	1.86	±1.14
B	1176.44	±21.64	16931.17	±3861.05	4040.46	2563.45	±459.02	8491.62	±1015.58
Si	762.41	±12.69	101591.13	±36545.65	29753.71	26526.52	±2852.99	38483.14	±8771.22
V	0.03	±0.002	252.84	±89.24	51.75	17.09	±5.25	44.44	±27.25
Rb	<0.01	±<0.001	10747.75	±2835.47	5695.06	10055.19	±7719.40	5612.63	±1503.61
Sr	0.03	±0.005	230.92	±129.98	261.05	263.04	±70.09	95.14	±56.46
Ba	0.04	±0.014	317.64	±248.72	82.31	165.39	±115.34	56.71	±33.44
Al	1.41	±0.13	2300.15	±2258.58 1	12.57	22.42	±15.48	22.52	±1.88
Pb	0.01	±0.002	2.73	±2.61	0.35	0.14	±0.02	0.03	±0.01
Hg	0.01	±<0.001	0.12	±0.07	0.03	0.04	±0.01	0.04	±0.02
Ti	<0.01	±<0.001	49.59	±47.22	0.53	0.60	±0.20	1.45	±1.08

Table 8. Mean (± standard error) elemental concentration in 15ml of filtrate (typically consumed per person on a daily basis) (n=7; n=2 at each study site except for Telela where n=1).

	Crop Ash Filtrate						
	Dog Abam		**Telela**	**Arok**		**Tit**	
Element	*(mg/15ml)*		*(mg/15ml)*	*(mg/15ml)*		*(mg/15ml)*	
Ca	1.78	±0.51	0.91	0.98	±0.04	0.99	±0.07
P	0.45	±0.15	0.04	0.08	±0.01	0.05	±0.009
K	199.13	±43.65	132.67	109.96	±47.40	214.53	±5.41
Na	0.71	±0.04	0.25	0.17	±0.004	0.25	±0.05
Mg	0.18	±0.18	2.59	2.50	±0.63	1.88	±1.87
Fe	0.006	±0.006	<0.001	<0.001	±<0.001	<0.001	±<0.001
Zn	0.001	±0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
Mn	0.002	±0.002	<0.001	<0.001	±<0.001	<0.001	±0.001
Cu	0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
Se	<0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
Mo	0.001	±0.001	0.003	0.001	±0.001	0.003	±0.002
Cr	0.003	±0.002	<0.001	<0.001	±<0.001	0.001	±0.001
Ni	<0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
Co	<0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
As	<0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
B	0.254	±0.06	0.061	0.038	±0.007	0.127	±0.02
Si	1.524	±0.55	0.446	0.398	±0.04	0.577	±0.13
V	0.004	±0.001	0.001	<0.001	±<0.001	0.001	±<0.001
Rb	0.161	±0.04	0.085	0.151	±0.12	0.084	±0.02
Sr	0.003	±0.002	0.004	0.004	±0.001	0.001	±0.001
Ba	0.005	±0.004	0.001	0.002	±0.002	0.001	±0.001
Al	0.035	±0.03	<0.001	<0.001	±<0.001	<0.001	±<0.001
Pb	<0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
Hg	<0.001	±<0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001
Ti	0.001	±0.001	<0.001	<0.001	±<0.001	<0.001	±<0.001

Table 9. Elemental contents of beans cooked in Plain (Control), Table Salt, Ground Salt and Ash Filtrate. The ash filtrate used was from the 2011 Dog Abam ash sample. The Dog Abam ash sample was used for the cooking trials to maintain congruence with the palatability trials. Values are expressed in oven-dried weight of beans.

	Treatment			
Element	**Control** *(mg/kg)*	**Table Salt** *(mg/kg)*	**Ground Salt** *(mg/kg)*	**Ash Filtrate** *(mg/kg)*
Ca	1355	1233	2168	1612
P	4317	5090	4469	4240
K	11936	14104	13360	12821
Na	579	10213	11274	653
Mg	1605	1851	1858	1563
Fe	57.11	69.99	202.73	67.83
Zn	30.86	31.59	25.59	31.11
Mn	18.55	19.50	26.10	23.79
Cu	10.81	9.65	9.37	10.60
Se	<0.01	0.18	<0.01	<0.01
Mo	1.92	1.49	1.75	1.77
Cr	0.26	0.38	0.89	0.30
Ni	2.31	2.21	2.01	1.71
Co	0.39	0.40	0.44	0.29
As	<0.01	<0.01	0.01	<0.01
B	93.66	84.70	83.66	99.49
Si	456.14	469.84	566.83	478.97
V	0.03	0.05	0.68	0.07
Rb	10.47	12.54	11.29	10.40
Sr	9.78	7.66	21.47	11.01
Ba	9.92	7.10	13.78	9.27
Al	76.43	85.22	219.52	86.55
Pb	0.02	0.02	0.09	0.03
Hg	<0.01	<0.01	<0.01	<0.01
Ti	1.02	1.98	42.22	1.75

Table 10. Daily intake of elements (mg) from an 80 g serving size for adults of air-dried beans cooked in Plain (Control), Table Salt, Ground Salt and Ash Filtrate and recommended daily intake (RDI) and upper limit (UL) amounts for Uganda (National Drug Authority 2009) and Canada (Health Canada 2010).

Element		Uganda*		Canada*		Treatment			
						Control	Table Salt	Ground Salt	Ash Filtrate
		RDI	UL	RDI	UL	(*mg*)	(*mg*)	(*mg*)	(*mg*)
Macro	**Ca**	1000	2000	1000	2500	104	95	167	124
	P	1000	2000	1000	2500	332	392	344	327
	K	3500	7000	4700	nd	919	1086	1029	987
	Na	2400	4800	1500	2300	45	786	868	50
	Mg	400	800	320/420	350**	124	142	143	120
Micro	**Fe**	8/18	36	8/18	45	4.40	5.39	15.61	5.22
	Zn	15	30	8/11	40	2.38	2.43	1.97	2.40
	Mn	2	4	1.8/2.3	11	1.43	1.50	2.01	1.83
	Cu	2	4	0.90	10	0.83	0.74	0.72	0.82
	Se	0.070	0.14	0.055	0.40	<0.01	0.01	<0.01	<0.01
	Mo	0.075	0.15	0.045	2	0.15	0.11	0.13	0.14
	Cr	0.20	0.40	0.030	nd	0.02	0.03	0.07	0.02
	Ni	nd	nd	nd	1	0.18	0.17	0.15	0.13
	Co	na	na	na	na	0.03	0.03	0.03	0.02
Trace	**As**	nd	nd	nd	nd	<0.01	<0.01	0.00	<0.01
	B	nd	3	nd	20	7.21	6.52	6.44	7.66
	Si	nd	nd	nd	nd	35.12	36.18	43.65	36.88
	V	nd	nd	nd	nd	0.00	0.00	0.05	0.01

Table 10. Continued.

Element		Uganda*		Canada*		Treatment			
						Control	Table Salt	Ground Salt	Ash Filtrate
		RDI	UL	RDI	UL	*(mg)*	*(mg)*	*(mg)*	*(mg)*
Unknown necessity or toxic	**Rb**	0	na	0	na	0.81	0.97	0.87	0.80
	Sr	0	0	0	0	0.75	0.59	1.65	0.85
	Ba	0	na	0	na	0.76	0.55	1.06	0.71
	Al	0	0	0	0	5.88	6.56	16.90	6.66
	Pb	0	0.243	0	0	0.00	0.00	0.01	0.00
	Hg	0	0	0	0	<0.01	<0.01	<0.01	<0.01
	Ti	na	na	na	na	0.08	0.15	3.25	0.14

na = not available; nd = lack of sufficient data.

*Recommended daily intake (RDI) and upper limit (UL) levels are for healthy, 19-50 year old individuals, differentiated for males/females if applicable.

**The upper limit for magnesium is for intake from a pharmacological source only and does not apply to intake from food or water.

6.4.2. Participant responses to ash filtrate use and health related questions

Interview responses (n=20) verified the crop species most commonly used for making ash filtrate in this region as *P. vulgaris* and *S. indicum*, with other crops used only infrequently due to availability (Figure 12). Crops are harvested annually, and the dry ash is stored for use throughout the year. Frequency of filtrate use varies between households from daily to almost daily (5-6 times per week). Use of filtrate is, 'always for use when cooking beans and some vegetables' (staple foods in this region). Most respondents felt no difference in well-being after consuming food cooked with ash filtrate, while a few said they felt better and happy, and only one said they got a chest pain (heartburn) after its consumption. Consistent with the previous answer, most women did not think that the use of ash filtrate use would have any effect on their health, and two thought that when the filtrate is over-added then there may be some adverse effect; "when you misadd the *kado atwona* [ash filtrate], you can feel some heartburn and makes me think it could be of some health problem". However, many women stated that if a practice was not healthy for them then a medical professional would have told them not to do it. Of the women interviewed, common health complaints included chest pain, heartburn, hypertension, ulcers, and stomach pain.

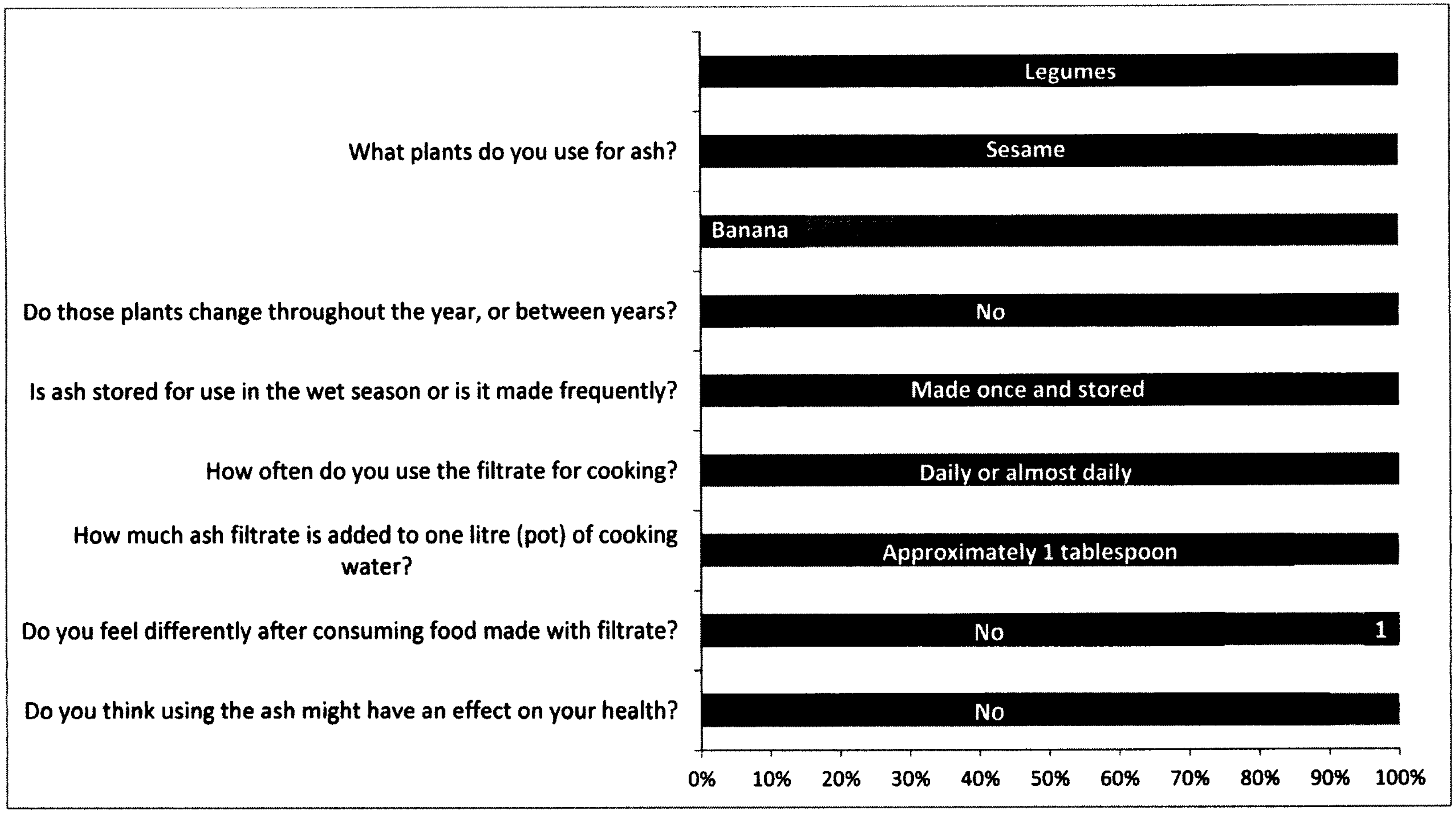

Figure 12. Summary of the responses of interview subjects to the utilisation and health related impacts of using ash filtrate to cook beans in Northern Uganda (n=20).

[1] Chest pain (heartburn) was reported by one respondent.

6.5. Discussion

Concentration of mineral levels found in the oven-dried crop ash for calcium, magnesium, potassium, iron, manganese, boron, strontium, barium and aluminum are quite high, and consistent with earlier findings (Calloway *et al.* 1974; Kuhnlein 1980; Kaputo 1996; Mamiro *et al.* 2011). Iron and aluminum are particularly abundant in the soils of the region (ISRIC 2013), and are easily taken up by plants in acidic soil conditions such as those in Northern Uganda (Brown 1978; Kochian, Pineros, & Hoekenga, 2005). As such, the elevated content of these elements in the ash was to be expected. There is likely further addition of elements into the ash from the inadvertent inclusion of soil; occurring when soil is left attached to the plant parts during harvest and when ash is scooped from the ground after the plant parts are burnt. Low concentration of sodium and high concentration of potassium in the plant ash, with the converse being true for combined salts, confirm previous findings (Wanjekeche *et al.* 2003; Mamiro *et al.* 2011). The high standard error for the combined salts reflects the crude nature of traditional salts and the much more uniform nature of the commercial salt. High potassium and sodium concentration are important in explaining the high alkalinity of the respective cooking treatments (Makanjuola & Beetlestone 1975; Mamiro *et al.* 2011).

The analysis of elemental transfer from ash to ash filtrate showed that generally, very low amounts of elements are being leached into the filtrate (Table 6), with a few exceptions (e.g., molybdenum and silicon). Subsequently, elemental concentration in the amount typically consumed per person per day (Table 8) is very low. The generation of filtrate for this study (the use of filter paper instead of ripped grass) may have decreased the elemental transfer from ash to filtrate due to low porosity (Horowitz *et al.* 1996). However, the color of filtrates from the same ash source (Dog Abam) created in both Uganda (with a spear grass filter bed) and Canada (with filter paper) was very similar (Figure 13).

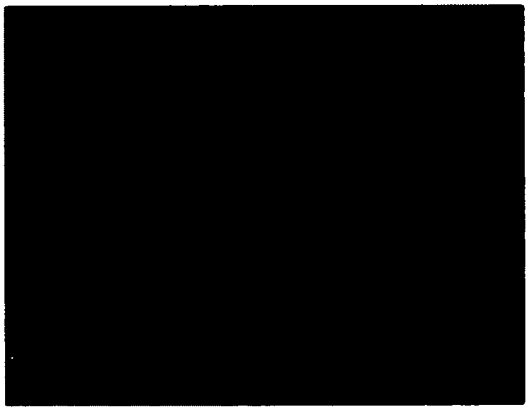

Figure 13. Ash filtrate samples created from Dog Abam crop ash in field (in cup) and in laboratory (in vial, inset).

While the colour of ash filtrate did vary by study site (where the ash was obtained), there was no apparent pattern to colour and elemental concentration. Reasons for variations in elemental levels among both dry ash and ash filtrate samples may be the result of using different varieties of *P. vulgaris*, or simply due to inherent variation within samples. Analysis of pH values for all filtrate samples proved them highly alkaline, confirming other results in this study as well as a previous finding (Kaputo 1996). Comparison of the pure ash filtrate alkalinity from Dog Abam (10.8 ± 0.1) with that of the cooking water prepared with ash filtrate (1.5% v/v) from Dog Abam (10.5 ± 0.3) shows very little dilution effect (Table 2, Chapter 4).

Legumes (beans) make up a significant percentage of the diet in Northern Uganda, and are eaten on a daily or almost daily basis. A typical daily serving size is approximately

80 g dried (240 g cooked) beans cooked with 15 ml filtrate per person, which are usually eaten over two to three meals throughout the day. Elemental contents in a 15 ml ash filtrate do not appear to be a health concern as they are well within the dietary reference intake amounts for required macro or micro elements by both Ugandan and Canadian standards (National Drug Authority 2009; Health Canada 2010). There are also no detectable levels of any heavy or toxic metals (e.g., mercury or lead) which could pose a health risk (Table 10). As the diet of people in rural Northern Uganda is generally quite limited, it is possible that the filtrate is actually providing a small amount of supplemental elements to their diet. For example, a typical daily consumption of filtrate could provide minor contributions between 100 mg and 200 mg of potassium. The use of ash filtrate as a supplement is similar to the practice of geophagy. Geophagy is the deliberate consumption of soil, which is often used as a type of curative traditional medicine, most often in developing countries (Abrahams 1997). It is often used as a source of iron and other trace minerals, where diet is limited.

Similar elemental concentrations of beans cooked in Type 1 water (control) and those cooked with ash filtrate reflect the low mineral input from filtrate (Table 9). In a typical daily portion size for adults (240 g of cooked beans), there are no elemental levels in the ash filtrate treatment of health concern (Table 10), and only the level of boron exceeds recommended daily intake (RDI) or upper limit (UL) set by Ugandan or Canadian guidelines. However, the level of boron in all treatments was comparable (including those cooked in plain water), and is therefore an inherent property of the beans themselves and not an issue being introduced by the cooking additive.

While the ash filtrate may be contributing small amounts of potentially beneficial elements to the diet, its pH level could be causing deleterious and counteractive nutritional effects (Mamiro *et al.* 2011). The filtrate created from ash from Dog Abam used to cook the bean sample has an alkaline pH of 10.8 ± 0.01, caused by the high content of potassium. Food additives which are highly alkaline have been shown to have various negative consequences on the nutritional aspects of cooked beans. Substantial decreases in bioavailability of iron (37%) and zinc (35%) have been observed with the use of plant ash (Mamiro *et al.* 2011), which may contribute to specific mineral deficiencies in the diet. Also of concern are the loss of nutrients, particularly thiamine (Sherman & Burton 1926; Onayemi *et al.* 1986; Kaputo 1996) and riboflavin (Kaputo 1996) and the destruction of essential amino acids (Minka *et al.* 1999) in the presence of high pH foods.

The ground salt treatment also had a high pH of 9.6 ± 0.06 (Chapter 4.4), which would raise the same concerns as those listed above. However, its alkalinity is brought about by its high level of sodium (Mamiro *et al.* 2011). Daily serving sizes of beans cooked with ground salt contain about one third the RDI of sodium for Ugandans, which is exacerbated by the addition of table salt to the sauce that beans are typically served with. These beans have a relatively high content of iron as well, which exceeds the RDI for men by both Ugandan and Canadian standards. Ground salt also contains high content of fluoride, an issue not seen with ash filtrate, which can lead to exceed the daily consumption limit (Mabelya *et al.* 1997; Nielsen 1999; Nielsen & Dahi 2002; Kaseva 2006). Given its potentially numerous negative effects, these results support previous findings which suggest ground salt should not be used as a cooking additive.

The complexity of factors that contribute to the possible health implications of plant ash filtrate consumption limit definitive conclusions within the context of this study. However, probable negative dietary impacts of its use have been identified. Further investigation of individuals in this area should be conducted to accurately identify elemental levels present through hair, blood and urine sampling, in addition to a more in-depth survey of current health status. These investigations could help to provide a conclusive stance on the nutritional effects of using ash filtrate in food preparation.

Chapter 7. General discussion

The intent of this research was to explore the use of crop ash filtrate as a cooking additive in rural Northern Uganda. The examination of practical, cultural, and mineralogical aspects of ash filtrate provided insight into this practice that is widely used but little studied. Based on the results of this study, it can be concluded that the use of ash filtrate has both positive functional purposes and probable negative dietary consequences.

The addition of ash filtrate from *P. vulgaris* crop residue to cooking water decreased required cooking time for hard-to-cook legumes confirming anecdotal beliefs about this practice. Ground salt also greatly decreased cooking time. The reduction of cooking time seen in both traditional treatments is due to the highly alkaline conditions created by high levels of potassium and sodium, respectively, which expedites ionization of pectic substances in the beans' cells and improves softening (Ankrah & Dovlo 1978; Uzogara *et al.* 1990; Onwuka & Okala 2003). This decrease in required cooking time of legumes has important unseen implications on fuel wood supply. If additives were not used to shorten cooking time there would be an even greater demand for fuel wood, translating into further deforestation.

Blind palatability tests did not support anecdotal beliefs that beans cooked with ash filtrate are the most preferred. In this study, table salt was not added to the cooked beans nor were the beans served with a sauce, which is a very common practice in Northern Uganda. It may be that the lack of added table salt and sauce with the beans influenced flavour preferences. The beans cooked in table salt and in ground salt were the most favored treatments. This may be the result of a higher content of sodium, as identified through elemental analysis.

In assessing implications of the use of ash filtrate as a cooking method on the potential health and well-being of people in this study, the results were not definitive. The highly alkaline nature of the filtrate could have deleterious effects on the mineral and nutritional content of beans cooked with the filtrate (Wanjekeche *et al.* 2003; Mamiro *et al.* 2011). Alternatively, the use of the ash filtrate may be providing small amounts of minerals to the people in this region who have a limited diet and few opportunities to access these minerals in other foods. However, possible negative impacts brought about by the pH characteristic of ash filtrate likely outweigh any elemental contribution it makes to the diet.

7.1 Implications and recommendations for future research

There may be several benefits to the use of plant ash filtrate as a cooking additive, including contribution to a cultural identity. However, the only clear benefit is the reduction in cooking time. Given the potentially detrimental nutritional aspects of consuming foods prepared with ash filtrate, I recommend that consideration be given to the use of an alternative method of decreasing cooking time for hard-to-cook legumes. Presoaking of dried legumes is a practice commonly used in other cultures, and may be the best method to replace the use of ash filtrate as it has been proven very effective in decreasing cooking time (Silva *et al.* 1981; Njoku *et al.* 1989; de León *et al.* 1992). This is an affordable and accessible method for use in rural areas. Educational programs raising awareness about the effects of using ash filtrate as a cooking additive and promoting presoaking as an alternative could be developed and implemented locally. A traditional practice that has practical as well as cultural significance is not likely to change quickly or easily. In sharing the results of this study with participants, there would be an opportunity to support local women to create change and carry it forward into their communities.

In this study, I provide the first comprehensive look into this indigenous practice through analysis of the elemental content of ash filtrate and the impact of cultural and traditional beliefs about the use of ash filtrate. While this work provides a foundation platform to study this and similar practices in other regions, financial and logistical constraints prevented a more in-depth analysis of health impacts. Further research should be conducted in Northern Uganda and other areas where this practice is in use, to determine the specific health effects of ash filtrate use. Such studies could include, but not limited to, biological analysis, detailed nutritional studies, and/or long-term monitoring of filtrate consumers.

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Appendix A. Semi-structured interview template

Code: ________

History

1. How long have you been doing this practice?
2. How long has your family been doing it?
3. Who did you learn this from?
4. Is this practice an important part of your life or culture? How so?

Materials and methods

5. What plants do you use for ash?
6. Do those plants change throughout the year, or between years?
7. Who usually helps in the ash making process?
8. Is ash stored for use in the wet season or is it made frequently?
9. How often do you use the filtrate for cooking?
10. How much kado atwona is added to a 1-litre pot of water? What would be the ratio of filtrate to water?

Reasoning

11. Does adding ash filtrate to the cooking water make food cook faster? How much faster?
12. Does adding ash filtrate to the cooking water make the food taste better?
13. Could regular salt be used instead (are ash and salt interchangeable)?
14. Do you sell your ash?

Perceptions

15. Do you feel differently after consuming food made with ash water?
16. Do you think using the ash might have an effect on your health?

Socioeconomic factors

17. Age
18. Education level
19. Number of members in family
20. Monthly household income
21. Health issues

Appendix B. Research assistant confidentiality agreement

Research Assistant Confidentiality Agreement

This study, "Use of filtrate from crop ash for cooking food in rural households in Northern Uganda", is being undertaken by Tara Bergeson at the University of Northern British Columbia.
The study has 4 objectives:

1. To compare cooking times for a common food (e.g., beans) in plain water (control) and water containing known concentrations of commercial salt, local rock salt, and ash filtrate (treatments).
2. To assess palatability of the common food cooked in each of the four treatments.
3. To assess whether specific socio-economic factors affect palatability preferences and use of the cooking practice.
4. To determine the chemical composition of the ash filtrate and to assess and identify potentially toxic or unhealthy constituents.

Data from this study will be used to inform a Master's Thesis.

I, ______________________________, agree to:

1. Keep all the research information shared with me confidential by not discussing or sharing the research information in any form or format (e.g. disks, tapes, transcripts) with anyone other than the Principal Investigator(s);
2. Keep all research information in any form or format secure while it is in my possession;
3. Return all research information in any form or format to the Principal Investigator(s) when I have completed the research tasks;
4. After consulting with the Principal Investigator(s), erase or destroy all research information in any form or format regarding this research project that is not returnable to the Principal Investigator(s) (e.g. information sorted on computer hard drive).

Research Assistant:

______________________	______________________	______________
(print name)	(signature)	(date)

Principal Investigator:

______________________ ______________________ ______________
(print name) (signature) (date)

If you have any questions or concerns about this study, please contact:

Dr. Chris Opio
Ecosystem Science and Management Program
University of Northern British Columbia
3333 University Way, Prince George , British Columbia, V2N 4Z9
000 1 (250) 960-5868
Email: opio@unbc.ca

This study has been reviewed by the Research Ethics Board at the University of Northern British Columbia. For questions regarding participants rights and ethical conduct of research, contact the Office of Research and Graduate Programs at: 000 1 (250) 960-6735, or by email at: reb@unbc.ca.

Appendix C. Participant consent form

Consent for Participation in Collection of Non-Identifying Information

Invitation to participate: You are invited to participate in this research because you live in Kamdini Parish, Oyam District, Uganda and are familiar with the use of ash filtrate for cooking.

Purpose and goals of study: The purpose of this study is to examine the practice of making and using crop ash filtrate for cooking in order to understand this practice.

Explanation of procedures: If you are a partial participant, you will complete a taste test survey of beans cooked in different water treatments. If you are a full participant, you will complete this taste test in addition to an interview and the procedures of ash filtrate creation with the Investigator and Research Assistant. The interview and ash creation procedures will be recorded with a voice and/or video recorder for future reference of the Investigator and Supervisor only.

Potential risks: There are no known expected risks or discomforts involved with your participation in this study.

Potential benefits: You will have the opportunity to learn about the research process and contribute to a study which may positively impact your communities' well-being.

Confidentiality of the data: Your name will not be associated in any way with your data. Your consent form and your data will be stored separately on digital memory devices for transport back to Canada where the devices will be kept at the University of Northern British Columbia in Room 8-314. The data will be destroyed/deleted after completion of this study, or approximately two years. Only the Investigator and Supervisor will have access to the data.

Withdrawal from the study: Participation is voluntary. There is no penalty for non-participation, or withdrawal from the study. If you decide to participate, you are free to withdraw your consent and to discontinue participation at any time without penalty. If you decide to withdraw, all data and information pertaining to you will be deleted.

Offer to answer questions: If you have any questions please feel free to ask. If you have any questions later on, you may contact the Investigator or Supervisor. If you would like to get a copy of the study results, the Northern Uganda Development Foundation (Manager Geoffrey Odongo) will be able to provide one once the study is complete. Thank you for your time and interest.

You are making a decision whether or not to participate. Your signature indicates that you have decided to participate having read or verbally understood the information above. You will be given a copy of this form to keep.

Participant Signature: ______________________________ **Date:** ____________________

Code: ________

Participant Signature: ______________________________ **Date:** ____________________

Investigator: Tara Bergeson (0777 609257) **Email: bergeson@unbc.ca**

Supervisor: Dr. C. Opio (000-1-250-960-5868) **Email: opio@unbc.ca**

NUDF Field Manager: Geoffrey Odongo (0777 172063) **Email: geoffrey@nudf.org**

If you have any complaints about the project, please feel free to contact the Office of Research at the University of Northern British Columbia (reb@unbc.ca or 000-1-250-960-6735).

Translation of form to the local dialect (Luo) will be done by the research assistant to ensure the participant understands in either Luo or English. The tone of the form may change due to differing views of politeness and terminology but the content will remain the same.

Appendix D. Palatability rating template

Code: _______

Village: _________________

Age: ___________________

Education level: __________

Occupation: _____________

Male ☐ Female ☐

Please circle the best numbered answer for each description of the cooked beans in each container.

Pot 1:

Colour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Smell	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Flavour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Texture	1	2	3	4	5
	Too soft		Neutral		Good
Texture	1	2	3	4	5
	Too hard		Neutral		Good
Bitterness	1	2	3	4	5
	Bitter/Chalky		Neutral		Acceptable
Overall Acceptability	1	2	3	4	5
	Extremely poor/ Unacceptable		Neutral		Extremely good/ Acceptable

Code: ___________

Pot 2:

Colour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Smell	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Flavour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Texture	1	2	3	4	5
	Too soft		Neutral		Good
Texture	1	2	3	4	5
	Too hard		Neutral		Good
Chalkiness	1	2	3	4	5
	Bitter/Chalky		Neutral		Acceptable
Overall Acceptability	1	2	3	4	5
	Extremely poor/ Unacceptable		Neutral		Extremely good/ Acceptable

Code: ___________

Pot 3:

Colour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Smell	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Flavour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Texture	1	2	3	4	5
	Too soft		Neutral		Good
Texture	1	2	3	4	5
	Too hard		Neutral		Good
Chalkiness	1	2	3	4	5
	Bitter/Chalky		Neutral		Acceptable
Overall Acceptability	1	2	3	4	5
	Extremely poor/ Unacceptable		Neutral		Extremely good/ Acceptable

Code: ___________

Pot 4:

Colour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Smell	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Flavour	1	2	3	4	5
	Extremely dislike		Neutral		Extremely like
Texture	1	2	3	4	5
	Too soft		Neutral		Good
Texture	1	2	3	4	5
	Too hard		Neutral		Good
Chalkiness	1	2	3	4	5
	Bitter/Chalky		Neutral		Acceptable
Overall Acceptability	1	2	3	4	5
	Extremely poor/ Unacceptable		Neutral		Extremely good/ Acceptable